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(54) Genomic DNA encoding a polypeptide capable of inducing the production of interferon-gamma

(57) Disclosed is a genomic DNA encoding a polypeptide capable of inducing the production of interferon-γ by immunocompetent cells. The genomic DNA efficiently expresses the polypeptide with high biological activities of such as inducing the production of interferon-γ by immunocompetent cells, enhancing killer cells' cytotoxicity and inducing killer cells' formation, when introduced into mammalian host cells. The high biological activities of the polypeptide facilitate its uses to treat and/or prevent malignant tumors, viral diseases, bacterial infectious diseases and immune diseases without serious side effects when administered to humans.

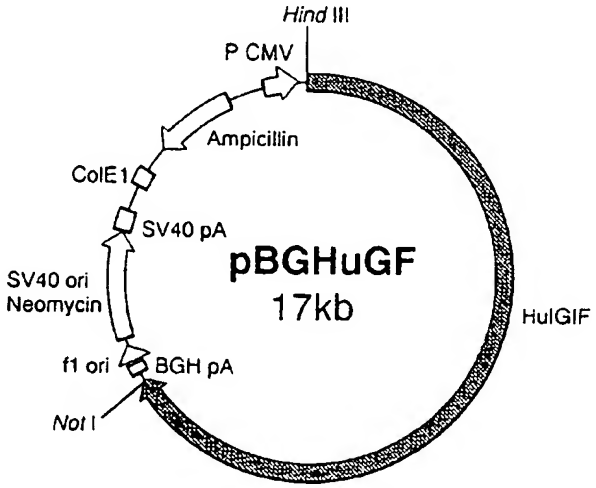

FIG.1

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Description

The present invention relates to a genomic DNA, more particularly, a genomic DNA encoding a polypeptide capable of inducing the production of interferon-γ (hereinafter abbreviated as "IFN-γ") by immunocompetent cells.

The present inventors successfully isolated a polypeptide capable of inducing the production of IFN-γ by immunocompetent cells and cloned a cDNA encoding the polypeptide, which is disclosed in Japanese Patent Kokai No. 27,189/96 and 193,098/96. Because the present polypeptide possesses the properties of enhancing killer cells' cytotoxicity and inducing killer cells' formation as well as inducing IFN-γ, a useful biologically active protein, it is expected to be widely used as an agent for viral diseases, microbial diseases, tumors and/or immunopathies, etc.

It is said that a polypeptide generated by a gene expression may be partially cleaved and/or glycosylated by processing with intracellular enzymes in human cells. A polypeptide to be used in therapeutic agents should be preferably processed similarly as in human cells, whereas human cell lines generally have a disadvantage of less producing the present polypeptide, as described in Japanese Patent Application No.269,105/96. Therefore, recombinant DNA techniques should be applied to obtain the present polypeptide in a desired amount. To produce the polypeptide processed similarly as in human cells using recombinant DNA techniques, mammalian cells should be used as the hosts.

In view of foregoing, the first object of the present invention is to provide a DNA which efficiently expresses the polypeptide production when introduced into a mammalian host cell.

The second object of the present invention is to provide a transformant into which the DNA is introduced.

The third object of the present invention is to provide a process for preparing a polypeptide, using the transformant.

[Means to Attain the Object]

The present inventors' energetic studies to attain the above objects succeeded in the finding that a genomic DNA encoding the present polypeptide efficiently expresses the polypeptide production when introduced into mammalian host cells. They found that the polypeptide thus obtained possessed significantly higher biological activities than that obtained by expressing a cDNA encoding the polypeptide in *Escherichia coli*.

The first object of the present invention is attained by a genomic DNA encoding a polypeptide with the amino acid sequence of SEQ ID NO:1 (where the symbol "Xaa" means "isoleucine" or "threonine") or its homologous one, which induces interferon-γ production by immunocompetent cells.

The second object of the present invention is attained by a transformant formed by introducing the genomic DNA into a mammalian host cell.

The third object of the present invention is attained by a process for preparing a polypeptide, which comprises (a) culturing the transformant in a nutrient medium, and (b) collecting the polypeptide from the resultant culture.

FIG.1 is a restriction map of a recombinant DNA containing a genomic DNA according to the present invention.

Explanation of the symbols are as follows: The symbol *"Hin* dIII" indicates a cleavage site by a restriction enzyme *Hin* dIII, and the symbol "HuIGIF" indicates a genomic DNA according to the present invention.

The followings are the preferred embodiments according to the present invention. This invention is made based on the identification of a genomic DNA encoding the polypeptide with the amino acid sequence of SEQ ID NO:1 or its homologous one, and the finding that the genomic DNA efficiently expresses the polypeptide with high biological activities when introduced into mammalian host cells. The genomic DNA of the present invention usually contains two or more exons, at least one of which possesses a part of or the whole of the nucleotide sequence of SEQ ID NO:2. The wording "a part" includes a nucleotide and a sequential nucleotides consisting of two or more nucleotides in SEQ ID NO:2. Examples of the exons are SEQ ID NOs:3 and 4. Human genomic DNA may contain additional exons with SEQ ID NOs:5 to 7. Since the present genomic DNA is derived from a mammalian genomic DNA, it contains introns, as a distinctive feature in mammalian genomic DNAs. The present genomic DNA usually has two or more introns such as SEQ ID NOs:8 to 12.

More particular examples of the present genomic DNA include DNAs with SEQ ID NOs:13 and 14 or complementary sequences thereunto. The DNAs with SEQ ID NOs:13 and 14 are substantially the same. The DNA with SEQ ID NO: 14 contains coding regions for a leader peptide, consisting of the nucleotides 15,607th-15,685th, 17,057th-17,068th and 20,452nd-20,468th, coding regions for the present polypeptide, consisting of the nucleotides 20,469th-20,586th, 21,921st-22,054th and 26,828th-27,046th, and regions as introns, consisting of the nucleotides 15,686th-17,056th, 17,069-20,451st, 20,587th-21,920th and 22,055th-26,827th. The genomic DNA with SEQ ID NO:13 is suitable for expressing the polypeptide in mammalian host cells.

Generally in this field, when artificially expressing a DNA encoding a polypeptide in a host, one or more nucleotides in a DNA may be replaced by different ones, and appropriate promoter(s) and/or enhancer(s) may be linked to the DNA to improve the expressing efficiency or the properties of the expressed polypeptide. The present genomic DNA can be altered similarly as above. Therefore, as far as not substantially changing in the biological activities of the expressed polypeptides, the present genomic DNA should include DNAs encoding functional equivalents of the

polypeptide, formed as follows: One or more nucleotides in SEQ ID NOs:3 to 14 are replaced by different ones, the untranslated regions and/or the coding region for a leader peptide in the 5'- and/or 3'-termini of SEQ ID NOs:3, 4, 5, 6, 7, 13 and 14 are deleted, and appropriate oligonucleotides are linked to either or both ends of SEQ ID NO:13.

The present genomic DNA includes general DNAs which are derived from a genome containing the nucleotide sequences as above, and it is not restricted to its sources or origins as far as it is once isolated from its original organisms. For example, the present genomic DNA can be obtained by chemically synthesizing based on SEQ ID NOs:2 to 14, or by isolating from a human genomic DNA. The isolation of the present genomic DNA from such a human genomic DNA comprises (a) isolating a genomic DNA from human cells by conventional methods, (b) screening the genomic DNA with probes or primers, which are chemically synthesized oligonucleotides with a part of or the whole of the nucleotide sequence of SEQ ID NO:2, and (c) collecting a DNA to which the probes or primers specifically hybridize. Once the present genomic DNA is obtained, it can be unlimitedly replicated by constructing a recombinant DNA with an autonomously replicable vector by conventional method and then introducing the recombinant DNA into an appropriate host such as a microorganism or an animal cell before culturing the transformant or by applying a PCR method.

The present genomic DNA is very useful in producing the polypeptide by recombinant DNA techniques since it efficiently expresses the polypeptide with high biological activities when introduced into mammalian host cells. The present invention further provides a process for preparing a polypeptide using a specific genomic DNA, comprising the steps of (a) culturing a transformant formed by introducing the present genomic DNA into mammalian host cells, and (b) collecting the polypeptide which induces IFN-γ production by immunocompetent cells from the resultant culture.

The following explains the process for preparing the polypeptide according to the present invention. The present genomic DNA is usually introduced into host cells in the form of a recombinant DNA. The recombinant DNA, comprising the present genomic DNA and an autonomously replicable vector, can be relatively easily prepared by conventional recombinant DNA techniques when the genomic DNA is available. The vectors, into which the present genomic DNA can be inserted, include plasmid vectors such as pcD, pcDL-SRα, pKY4, pCDM8, pCEV4 and pME18S. The autonomously replicable vectors usually further contain appropriate nucleotide sequences for the expression of the present recombinant DNA in each host cell, which include sequences for promoters, enhancers, replication origins, transcription termination sites, splicing sequences and/or selective markers. Heat shock protein promoters or IFN-a promoters, as disclosed in Japanese Patent Kokai No.163,368/95 by the same applicant of this invention, enables to artificially regulate the present genomic DNA expression by external stimuli.

To insert the present genomic DNA into vectors, conventional methods used in this field can be arbitrarily used: Genes containing the present genomic DNA and autonomously replicable vectors are cleaved with restriction enzymes and/or ultrasonic, and the resultant DNA fragments and the resultant vector fragments are ligated. To cleave genes and vectors by restriction enzymes, which specifically act on nucleotides, more particularly, *Acc*I, *Bam*HI, *Bgl*I, *Bst*XI, *Eco*RI, *Hin*dIII, *Not*I, *Pst*I, *Sac*I, *Sal*I, *Sma*I, *Spe*I, *Xba*I, *Xho*I, etc., facilitate the ligation of the DNA fragments and the vector fragments. To ligate the DNA fragments and the vector fragments, they are, if necessary, first annealed, then treated with a DNA ligase *in vivo* or *in vitro*. The recombinant DNAs thus obtained can be unlimitedly replicated in hosts derived from microorganisms or animals.

Any cells conventionally used as hosts in this field can be used as the host cells: Examples of such are epithelial, interstitial and hemopoietic cells, derived from human, monkey, mouse and hamster, more particularly, 3T3 cells, C127 cells, CHO cells, CV-1 cells, COS cells, HeLa cells, MOP cells and their mutants. Cells which inherently produce the present polypeptide also can be used as the host cells: Example of such are human hemopoietic cells such as lymphoblasts, lymphocytes, monoblasts, monocytes, myeloblasts, myelocytes, granulocytes and macrophages, and human epithelial and interstitial cells derived from solid tumors such as pulmonary carcinoma, large bowel cancer and colon cancer. More particular examples of the latter hemopoietic cells are leukemia cell lines such as HBL-38 cells, HL-60 cells ATCC CCL240, K-562 cells ATCC CCL243, KG-1 cells ATCC CCL246, Mo cells ATCC CRL8066, THP-1 cells ATCC TIB202, U-937 cells ATCC CRL1593.2, described by J. Minowada et al. in *"Cancer Research"*, Vol.10, pp. 1-18 (1988), derived from leukemias or lymphoma including myelogenous leukemias, promyelocytic leukemias, monocytic leukemias, adult T-cell leukemias and hairy cell leukemias, and their mutants. The present polypeptide-processibility of these leukemia cell lines and their mutants is so distinguished that they can easily yield the polypeptide with higher biological activities when used as hosts.

To introduce the present DNA into the hosts, conventional methods such as DEAE-dextran method, calcium phosphate transfection method, electroporation method, lipofection method, microinjection method, and viral infection method as using retrovirus, adenovirus, herpesvirus and vaccinia virus, can be used. The polypeptide-producing clones in the transformants can be selected by applying the colony hybridization method or by observing the polypeptide production after culturing the transformants in culture media. For example, the recombinant DNA techniques using mammalian cells as hosts are detailed in *"Jikken-Igaku-Bessatsu Saibo-Kogaku Handbook* (The handbook for the cell engineering)" (1992), edited by Toshio KUROKI, Masaru TANIGUCHI and Mitsuo OSHIMURA, published by YODOSHA. CO., LTD., Tokyo, Japan, and *"Jikken-Igaku-Bessatsu Biomanual Series 3 Idenshi Cloning Jikken-Ho* (The experimen-

tal methods for the gene cloning)" (1993), edited by Takahi YOKOTA and Ken-ichi ARAI, published by YODOSHA CO., LTD., Tokyo, Japan.

The transformants thus obtained secrete the present polypeptide intracellularly and/or extracellularly when cultured in culture media. As the culture media, conventional ones used for mammalian cells can be used. The culture media generally comprise (a) buffers as a base, (b) inorganic ions such as sodium ion, potassium ion, calcium ion, phosphoric ion and chloric ion, (c) micronutrients, carbon sources, nitrogen sources, amino acids and vitamins, which are added depending on the metabolic ability of the cells, and (d) sera, hormones, cell growth factors and cell adhesion factors, which are added if necessary. Examples of individual media include 199 medium, DMEM medium, Ham's F12 medium, IMDM medium, MCDB 104 medium, MCDB 153 medium, MEM medium, RD medium, RITC 80-7 medium, RPMI-1630 medium, RPMI-1640 medium and WAJC 404 medium. The cultures containing the present polypeptide are obtainable by inoculating the transformants into the culture media to give a cell density of 1×10^4 - 1×10^7 cells/ml, more preferably, 1×10^5 - 1×10^6 cells/ml, and then subjecting to suspension- or monolayer-cultures at about 37°C for 1-7 days, more preferably, 2-4 days, while appropriately replacing the culture media with a fresh preparation of the culture media. The cultures thus obtained usually contain the present polypeptide in a concentration of about 1-100 μg/ml, which may vary depending on the types of the transformants or the culture conditions used.

While the cultures thus obtained can be used intact as an IFN-γ inducer, they are usually subjected to a step for separating the present polypeptide from the cells or the cell debris using filtration, centrifugation, etc. before use, which may follow a step for disrupting the cells with supersonication, cell-lytic enzymes and/or detergents if desired, and to a step for purifying the polypeptide. The cultures from which the cells or cell debris are removed are usually subjected to conventional methods used in this field for purifying biologically active polypeptides, such as salting-out, dialysis, filtration, concentration, separatory sedimentation, ion-exchange chromatography, gel filtration chromatography, adsorption chromatography, chromatofocusing, hydrophobic chromatography, reversed phase chromatography, affinity chromatography, gel electrophoresis and/or isoelectric focusing. The resultant purified polypeptide can be concentrated and/or lyophilized into liquids or solids depending on final uses. The monoclonal antibodies disclosed in Japanese Patent Kokai No.231,598/96 by the same applicant of this invention are extremely useful to purify the present polypeptide. Immunoaffinity chromatography using monoclonal antibodies yields the present polypeptide in a relatively high purity at the lowest costs and labors.

The polypeptide obtainable by the process according to the present invention exerts strong effects in the treatment and/or the prevention for IFN-γ- and/or killer cell-susceptive diseases since it possesses the properties of enhancing killer cells' cytotoxicity and inducing killer cells' formation as well as inducing IFN-γ, a useful biologically active protein, as described above. The polypeptide according to the present invention has a high activity of inducing IFN-γ, and this enables a desired amount of IFN-γ production with only a small amount. The polypeptide is so low toxic that it scarcely causes serious side effects even when administered in a relatively-high dose. Therefore, the polypeptide has an advantage that it can readily induce IFN-γ in a desired amount without strictly controlling the dosage. The uses as agents for susceptive diseases are detailed in Japanese Patent Application No.28,722/96 by the same applicant of this invention.

The present genomic DNA is also useful for so-called "gene therapy". According to conventional gene therapy, the present DNA can be introduced into patients with IFN-γ- and/or killer cell-susceptive diseases by directly injecting after the DNA is inserted into vectors derived from viruses such as retrovirus, adenovirus and adeno-associated virus or is incorporated into cationic- or membrane fusible-liposomes, or by self-transplanting lymphocytes which are collected from patients before the DNA is introduced. In adoptive immunotherapy with gene therapy, the present DNA is introduced into effector cells similarly as in conventional gene therapy. This can enhance the cytotoxicity of the effector cells to tumor cells, resulting in improvement of the adoptive immunotherapy. In tumor vaccine therapy with gene therapy, tumor cells from patients, into which the present genomic DNA is introduced similarly as in conventional gene therapy, are self-transplanted after proliferated *ex vivo* up to give a desired cell number. The transplanted tumor cells act as vaccines in the patients to exert a strong antitumor immunity specifically to antigens. Thus, the present genomic DNA exhibits considerable effects in gene therapy for diseases including viral diseases, microbial diseases, malignant tumors and immunopathies. The general procedures for gene therapy are detailed in *"Jikken-Igaku-Bessatsu Biomanual UP Series Idenshichiryo-no-Kisogijutsu* (Basic techniques for the gene therapy)" (1996), edited by Takashi ODAJIMA, Izumi SAITO and Keiya OZAWA, published by YODOSHA CO., LTD., Tokyo, Japan.

The following examples explain the present invention, and the techniques used therein are conventional ones used in this field: For example, the techniques are described in *"Jikken-Igaku-Bessatsu Saibo-Kogaku Handbook* (The handbook for the cell engineering)", (1992), edited by Toshio KUROKI, Masaru TANIGUCHI and Mitsuo OSHIMURA, published by YODOSHA CO., LTD., Tokyo, Japan, and *"Jikken-Igaku-Bessatsu Biomanual Series 3 Idenshi Clonong Jikken-Ho* (The experimental methods for the gene cloning)" (1993), edited by Takahi YOKOTA and Ken-ichi ARAI, published by YODOSHA CO., LTD., Tokyo, Japan.

Example 1

Cloning genomic DNA and determination of nucleotide sequence

Example 1-1

Determination of partial nucleotide sequence

Five ng of "PromoterFinder™ DNA PvuII LIBRARY", a human placental genomic DNA library commercialized by CLONTECH Laboratories, Inc., California, USA, 5 µl of 10 x Tth PCR reaction solution, 2.2 µl of 25 mM magnesium acetate, 4 µl of 2.5 mM dNTP-mixed solution, one µl of the mixed solution of 2 unit/µl rTth DNA polymerase XL and 2.2 µg/µl Tth Start Antibody in a ratio of 4:1 by volume, 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-CCATCCTAATACGACTCACTATAGGGC-3' as an adaptor primer, and 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-TTCCTCTTCCCGAAGCTGTGTAGACTGC-3' as an anti-sense primer, which was chemically synthesized based on the sequence of the nucleotides 88th-115th in SEQ ID NO:2, were mixed and volumed up to 50 µl with sterilized distilled water. After incubating at 94°C for one min, the mixture was subjected to 7 cycles of incubations at 94°C for 25 sec and at 72°C for 4 min, followed by 32 cycles of incubations at 94°C for 25 sec at 67°C for 4 min to perform PCR.

The reaction mixture was diluted by 100 folds with sterilized distilled water. One µl of the dilution, 5 µl of 10 x Tth PCR reaction solution, 2.2 µl of 25 mM magnesium acetate, 4 µl of 2.5 mM dNTP-mixed solution, one µl of the mixed solution of 2 unit/µl rTth DNA polymerase XL and 2.2 µg/µl Tth Start Antibody in a ratio of 4:1 by volume, 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-CTATAGGGCACGCGTGGT-3' as a nested primer, and 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-TTCCTCTTCCCGAAGCTGTGTAGACTGC-3' as an anti-sense primer, which was chemically synthesized similarly as above, were mixed and volumed up to 50 µl with sterilized distilled water. After incubating at 94°C for one min, the mixture was subjected to 5 cycles of incubations at 94°C for 25 sec and at 72°C for 4 min, followed by 22 cycles of incubations at 94°C for 25 sec and at 67°C for 4 min to perform PCR for amplifying a DNA fragment of the present genomic DNA. The genomic DNA library and reagents for PCR used above were mainly from "PromoterFinder™ DNA WALKING KITS", commercialized by CLONTECH Laboratories, Inc., California, USA

An adequate amount of the PCR product thus obtained was mixed with 50 ng of "pT7 Blue(R)", a plasmid vector commercialized by Novagen, Inc., WI, USA, and an adequate amount of T4 DNA ligase, and 100 mM ATP was added to give a final concentration of one mM, followed by incubating at 16°C for 18 hr to insert the DNA fragment into the plasmid vector. The obtained recombinant DNA was introduced into an *Escherichia coli* JM109 strain by the competent cell method to form a transformant, which was then inoculated into L-broth medium (pH 7.2) containing 50 µg/ml ampicillin and cultured at 37°C for 18 hr. The cells were isolated from the resulting culture, and then subjected to the conventional alkali-SDS method to collect a recombinant DNA. The dideoxy method analysis confirmed that the recombinant DNA contained the DNA fragment with a sequence of the nucleotides 5,150th-6,709th in SEQ ID NO:14.

Example 1-2

Determination of partial nucleotide sequence

PCR was performed in the same conditions as the first PCR in Example 1-1, but an oligonucleotide with the nucleotide sequence of 5'-GTAAGTTTTCACCTTCCAACTGTAGAGTCC-3', which was chemically synthesized based on the nucleotide sequence of the DNA fragment in Example 1-1, was used as an anti-sense primer.

The reaction mixture was diluted by 100 folds with sterilized distilled water. One µl of the dilution was placed into a reaction tube, and PCR was performed in the same conditions as used in the second PCR in Example 1-1 to amplify another DNA fragment of the present genomic DNA, but an oligonucleotide with the nucleotide sequence of 5'-GGGATCAAGTAGTGATCAGAAGCAGCACAC-3', which was chemically synthesized based on the nucleotide sequence of the DNA fragment in Example 1-1, was used as an anti-sense primer.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 1st-5,228th in SEQ ID NO:14.

Example 1-3

Determination of partial nucleotide sequence

0.5 µg of a human placental genomic DNA, commercialized by CLONTECH Laboratories, Inc., California, USA, 5 µl of 10 x PCR reaction solution, 8 µl of 2.5 mM dNTP-mixed solution, one µl of the mixed solution of 5 unit/µl "TAKARA LA Taq POLYMERASE" and 1.1 µg/µl "TaqStart ANTIBODY" in a ratio of 1:1 by volume, both of them are commercialized by Takara Syuzo Co., Tokyo, Japan, 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-CCTGGCTGCCAACTCTGGCTGCTAAAGCGG-3' as a sense primer, chemically synthesized based on a sequence of the nucleotides 46th-75th in SEQ ID NO:2, and 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-GTATTGTCAATAAATTTCATTGCCACAAAGTTG-3' as an anti-sense primer, chemically synthesized based on a sequence of the nucleotides 210th-242nd in SEQ ID NO:2, were mixed and volumed up to 50 µl with sterilized distilled water. After incubating at 94°C for one min, the mixture was subjected to 5 cycles of incubations at 98°C for 20 sec and at 68°C for 10 min, followed by 25 cycles of incubations at 98°C for 20 sec and 68°C for 10 min, with adding 5 sec in times to every cycle, and finally incubated at 72°C for 10 min to amplify further DNA fragment of the present genomic DNA. The reagents for PCR used above were mainly from "TAKARA LA PCR KIT VERSION 2", commercialized by Takara Syuzo Co., Tokyo, Japan.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 6,640th-15,671st in SEQ ID NO:14.

Experiment 1-4

Determination of partial nucleotide sequence

PCR was performed in the same conditions as the PCR in Example 1-3 to amplify further another DNA fragment of the present genomic DNA; but an oligonucleotide with the nucleotide sequence of 5'-AAGATGGCTGCTGAACCAGTAGAAGACAATTGC-3', chemically synthesized based on a sequence of the nucleotide 175th-207th in SEQ ID NO: 2, was used as a sense primer, an oligonucleotide with the nucleotide sequence of 5'-TCCTTGGTCAATGAAGAGAACTTGGTC-3', chemically synthesized based on a sequence of nucleotides 334th-360th in the SEQ ID NO:2, was used as an anti-sense primer, and after incubating at 98°C for 20 sec, the reaction mixture was subjected to 30 cycles of incubations at 98°C for 20 sec and at 68°C for 3 min, followed by incubating at 72°C for 10 min.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 15,604th-20,543rd in SEQ ID NO:14.

Example 1-5

Determination of partial nucleotide sequence

PCR was performed in the same conditions as the PCR in Example 1-4 to amplify further another DNA fragment of the present genomic DNA, but an oligonucleotide with the nucleotide sequence of 5'-CCTGGAATCAGATTACTTTGGCAAGCTTGAATC-3', chemically synthesized based on the sequence of the nucleotide 273rd-305th in SEQ ID NO: 2, was used as a sense primer, and an oligonucleotide with the nucleotide sequence of 5'-GGAAATAATTTTGTTCTCACAGGAGAGAGTTG-3', chemically synthesized based on the sequence of nucleotides 500th-531st in the SEQ ID NO:2, was used as an anti-sense primer.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 20,456th-22,048th in SEQ ID NO:14.

Example 1-6

Determination of partial nucleotide sequence

PCR was performed in the same conditions as the PCR in Example 1-4 to amplify further another DNA fragment

of the present genomic DNA, but an oligonucleotide with the nucleotide sequence of 5'-GCCAGCCTAGAGGTATGGCTGTAACTATCTC-3', chemically synthesized based on the sequence of the nucleotide 449th-479th in SEQ ID NO:2, was used as a sense primer, and an oligonucleotide with the nucleotide sequence of 5'-GGCATGAAATTTTAATAGCTAGTCTTCGTTTTG-3', chemically synthesized based on the sequence of nucleotides 745th-777th in the SEQ ID NO:2, was used as an anti-sense primer.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 21,996th-27,067th in SEQ ID NO:14.

Example 1-7

Determination of partial nucleotide sequence

PCR was performed in the same conditions as the first PCR in Example 1-2 to amplify further another DNA fragment in the present genomic DNA, but an oligonucleotide with the nucleotide sequence of 5'-GTGACATCATATTCTTTCAGAGAAGTGTCC-3', chemically synthesized based on the sequence of the nucleotide 575th-604th in SEQ ID NO:2, was used as a sense primer.

The reaction mixture was diluted by 100 folds with sterilized distilled water. One µl of the dilution was placed into a reaction tube, and PCR was performed in the same conditions as the second PCR in Example 1-2 to amplify further another DNA fragment of the present genomic DNA, but an oligonucleotide with the sequence of 5'-GCAATTTGAATCTTCATCATACGAAGGATAC-3', chemically synthesized based on a sequence of the nucleotides 624th-654th in SEQ ID NO:2, was used as a sense primer.

The DNA fragment was inserted into the plasmid vector similarly as in Example 1-1 to obtain a recombinant DNA. The recombinant DNA was replicated in *Escherichia coli* before being collected. The analysis of the collected recombinant DNA confirmed that it contained the DNA fragment with a sequence of the nucleotides 26,914th-28,994th in SEQ ID NO:14.

Example 1-8

Determination of complete nucleotide sequence

Comparing the nucleotide sequence of SEQ ID NO:2, which was proved to encode the present polypeptide, as disclosed in Japanese Patent Kokai No.193,098/96 by the same applicant of this invention, with the partial nucleotide sequences identified in Examples 1-1 to 1-7, it was proved that the present genomic DNA contained the nucleotide sequence of SEQ ID NO:14. SEQ ID NO:14, consisting of 28,994 base pairs (bp), was extremely longer than the SEQ ID NO:2, consisting of only 471 bp. This suggested that SEQ ID NO:14 contained introns, a characteristic of eukalyotic cells.

It was examined where partial nucleotide sequences of SEQ ID NO:2, i.e., exons, and the donor and acceptor sites in introns, respectively consisting of the nucleotides of GT and AG, located in SEQ ID NO:14. Consequently, it was proved that SEQ ID NO:14 contained at least 5 introns, which located in the order of SEQ ID NOs:10, 11, 12, 8 and 9 in the direction from the 5'- to the 3'-termini. Therefore, the sequences between the neighboring introns must be exons, which were thought to be located in the order of SEQ ID NOs:5, 6, 3, 4 and 7 in the direction from the 5'- to the 3'-termini. It was also proved that SEQ ID NO:7 contained the 3'-untranslated region other than the exons. The features of the sequence elucidated as this are arranged in SEQ ID NO:14.

As disclosed in Japanese Patent Application No. 269,105/96 by the same applicant of this invention, the present polypeptide is produced as a polypeptide with N-terminal amino acid of tyrosine other than methionine in human cells, which is observed in SEQ ID NO:1. This suggests that the present genomic DNA contains a leader peptide region in the upstream of the 5'-terminus of the present polypeptide-encoding region. A sequence consisting of 36 amino acids encoded by the upstream of the nucleotides 20,469th-20,471st. which is the nucleotides of TAC, are described as a leader peptide in SEQ ID NO:14.

Example 2

Preparation of recombinant DNA pBGHuGF for expression

0.06 ng of the DNA fragment in Example 1-4 in a concentration of 3 ng/50 µl, 0.02 ng of the DNA fragment, obtained by the methods in Example 1-5, 5 µl of 10 x LA PCR reaction solution, 8 µl of 2.5 mM dNTP-mixed solution, one µl of

the mixed solution of 5 unit/μl TAKARA LA Taq polymerase and 1.1 μg/μl TaqStart Antibody in a ratio of 1:1 by volume, 10 pmol of an oligonucleotide with the sequence of 5'-TCCGAAGCTTAAGATGGCTGCTGAACCAGTA-3' as a sense primer, chemically synthesized based on the nucleotide sequence of the DNA fragment in Example 1-4, and 10 pmol of an oligonucleotide with the nucleotide sequence of 5'-GGAAATAATTTTGTTCTCACAGGAGAGAGTTG-3' as an anti-sense primer, chemically synthesized based on the nucleotide sequence of the DNA fragment in Example 1-5, were mixed and volumed up to 50 μl with sterilized distilled water. After incubating at 94°C for one min, the mixture was subjected to 5 cycles of incubations at 98°C for 20 sec and at 72°C for 7 min, followed by 25 cycles of incubations at 98°C for 20 sec and 68°C for 7 min to perform PCR. The reaction mixture was cleaved by restriction enzymes *Hin*dIII and *Sph*I to obtain a DNA fragment of about 5,900 bp, with cleavage sites by HindIII and *Sph*I in its both termini.

PCR was performed in the same condition as above, but 0.02 ng of the DNA fragment in Example 1-5, 0.06 ng of the DNA fragment obtained in Example 1-6, an oligonucleotide with the nucleotide sequence of 5'-ATGTAGCG-GCCGCGGCATGAAATTTTAATAGCTAGTC-3' as an anti-sense primer, chemically synthesized based on the nucleotide sequence of the DNA fragment in Example 1-6, and an oligonucleotide with the sequence of 5'-CCTGGAATCA-GATTACTTTGGCAAGCTTGAATC-3' as a sense primer, chemically synthesized based on the DNA fragment in Example 1-6, were used. The reaction mixture was cleaved by restriction enzymes *Not*I and *Sph*I to obtain a DNA fragment of about 5,600 bp, with cleavage sites by *Not*I and *Sph*I in its both termini.

A plasmid vector "pRc/CMV", containing a *cytomegalovirus* promoter, commercialized by Invitrogen Corporation, San Diego, USA, was cleaved by restriction enzymes *Hin*dIII and *Not*I to obtain a vector fragment of about 5,500 bp. The vector fragment was mixed with the above two DNA fragments of about 5,900 bp and 5,600 bp, and reacted with T4 DNA ligase to insert the two DNA fragments into the plasmid vector. An *Escherichia coli* JM109 strain was transformed with the obtained recombinant DNA, and the transformant with the plasmid vector was selected by the colony hybridization method. The selected recombinant DNA was named as "pBGHuGF". As shown in FIG.1, the present genomic DNA, with the nucleotide sequence of SEQ ID NO:13, was ligated in the downstream of the cleavage site by the restriction enzyme HindIII in the recombinant DNA.

Example 3

Preparation of transformant using CHO cell as host

CHO-K1 cells ATCC CCL61 were inoculated into Ham's F12 medium (pH 7.2) containing 10 v/v % bovine fetal serum and proliferated by conventional manner. The proliferated cells were collected and washed with phosphate-buffered saline (hereinafter abbreviated as "PBS") followed by suspending in PBS to give a cell density of 1 x 10^7 cells/ml.

10 μg of the recombinant DNA pBGHuGF in Example 2 and 0.8 ml of the above cell suspension were placed in a cuvette and ice-chilled for 10 min. The cuvette was installed in "GENE PULSER", an electroporation device commercialized by Bio-Rad Laboratories Inc., Brussels, Belgium, and then pulsed once with an electric discharge. After pulsing, the cuvette was immediately took out and ice-chilled for 10 min. The cell suspension from the cuvette was inoculated into Ham's F12 medium (pH 7.2) containing 10 v/v % bovine fetal serum and cultured under an ambient condition of 5 v/v % CO_2 at 37°C for 3 days. To the culture medium was added G-418 to give a final concentration of 400 μg/ml, and the culturing was continued further 3 weeks under the same conditions. From abut 100 colonies formed, 48 colonies were selected, and a portion of each was inoculated into a well of culturing plates with Ham's F12 medium (pH7.2) containing 400 μg/ml G-418 and 10 v/v % bovine fetal serum and cultured similarly as above. Thereafter, to each well of the culturing plates was added 10 mM Tris-HCl buffer (pH 8.5) containing 5.1 mM magnesium chloride, 0.5 w/v % sodium deoxycholate, 1 w/v % NONIDET P-40, 10 μg/ml aprotinin and 0.1 w/v % SDS to lyse the cells.

50 μl aliquot of the cell lysates was mixed with one ml of glycerol and incubated at 37°C for one hour, before the polypeptides in the cell lysates were separated by the SDS-polyacrylamide gel electrophoresis. The separated polypeptides were transferred to a nitrocellulose membrane in usual manner, and the membrane was soaked in the culture supernatant of the hybridoma H-1, disclosed in Japanese Patent Kokai No.231,598/96 by the same applicant of this invention, followed by washing with 50 mM Tris-HCl buffer containing 0.05 v/v % TWEEN 20 to remove an excessive mount of the monoclonal antibody. Thereafter, the nitrocellulose membrane was soaked in PBS containing rabbit-derived anti-mouse immunoglobulin antibody for one hr, which was labeled with horseradish peroxidase, followed by washing 50 mM Tris-HCl buffer (pH 7.5) containing 0.05 v/v % TWEEN 20 and soaking in 50 mM Tris-HCl buffer (pH 7.5) containing 0.005 v/v % hydrogen peroxide and 0.3 mg/ml diaminobenzidine to develop colorations. The clone, which highly produced the polypeptide, was selected based on the color development and named "BGHuGF".

Example 4

Production of polypeptide by transformant and its physicochemical properties

The transformant BGHuGF in Experiment 3 was inoculated into Ham's F12 medium (pH 7.2) containing 400 μg/ml G-418 and 10 v/v % bovine fetal serum, and cultured under an ambient condition of 5 v/v % CO_2 at 37°C for one week. The proliferated cells were collected, washed with PBS, and then washing with 10-fold volumes of ice-chilled 20 mM Hepes buffer (pH 7.4), containing 10 mM potassium chloride and 0.1 mM ethylendiaminetetraacetate bisodium salt, according to the method described in *"Proceedings of The National Academy of The Sciences of The USA"*, vol. 86, pp.5,227-5,231 (1989), by M. J. Kostura et al. The cells thus obtained were allowed to stand in 3-fold volumes of a fresh preparation of the same buffer under an ice-chilling condition for 20 min and freezed at -80°C, succeeded by thawing to disrupt the cells. The resulting cells were centrifuged to collect the supernatant.

In parallel, THP-1 cells ATCC TIB 202, derived from a human acute monocytic leukemia, was similarly cultured and disrupted. Supernatant, obtained by centrifuging the resulting cells, was mixed with the supernatant obtained from the transformant BGHuGF and incubated at 37°C for 3 hr to react. The reaction mixture was applied to a column with "DEAE-SEPHAROSE", a gel for ion-exchange chromatography, commercialized by Pharmacia LKB Biotechnology AB, Upsalla, Sweden, equilibrated with 10 mM phosphate buffer (pH 6.6) before use. After washing the column with 10 mM phosphate buffer (pH 6.6), 10 mM phosphate buffer (pH 6.6) with a stepwise gradient of NaCl increasing from 0 M to 0.5 M was fed to the column, and fractions eluted by about 0.2 M NaCl were collected. The fractions were dialyzed against 10 mM phosphate buffer (pH 6.8) before applied to a column with "DEAE 5PW", a gel for ion-exchange chromatography, commercialized by TOSOH Corporation, Tokyo, Japan. To the column was fed 10 mM phosphate buffer (pH 6.8) with a linear gradient of NaCl increasing from 0 M to 0.5 M, and fractions eluted by about 0.2-0.3 M NaCl were collected.

While the obtained fractions were pooled and dialyzed against PBS, a gel for immunoaffinity chromatography with the monoclonal antibody were prepared according to the method disclosed in Japanese Patent Kokai No.231,598/96 by the same applicant of this invention. After the gel were charged into a plastic column and washed with PBS, the above dialyzed solution was applied to the column. To the column was fed 100 mM glycine-HCl buffer (pH 2.5), and the eluted fractions, which contained a polypeptide capable of inducing the production of IFN-γ by immunocompetent cells, were collected. After the collected fractions were dialyzed against sterilized distilled water and concentrated with a membrane filtration, the resultant was lyophilized to obtain a purified solid polypeptide in a yield of about 15 mg/l-culture.

Example for Reference

Expression in Escherichia coli

As disclosed in Japanese Patent Kokai No.193,098/96, a transformant pKHuGF which was obtained by introducing a cDNA with the nucleotide sequence of SEQ ID NO:2 into Escherichia coli as a host, was inoculated into L-broth medium containing 50 μg/ml ampicillin and cultured at 37°C for 18 hr under shaking conditions. The cells were collected by centrifuging the resulting culture, and then suspended in a mixture solution (pH 7.2) of 139 mM NaCl, 7 mM NaH_2PO_4 and 3 mM Na_2HPO_4, followed by supersonicating to disrupt the cells. After the cell disruptants were centrifuged, the supernatant was subjected to purifying steps similarly as in Example 4-1 to obtain a purified solid polypeptide in a yield of about 5 mg/l-culture.

Comparing the yields of the polypeptides in Example for Reference and in Example 4-1 shows that the use of a transformant, which is formed by introducing a genomic DNA encoding the present polypeptide into a mammalian cell as a host, strongly elevates the yield of the polypeptide per culture.

Example 4-2

Physicochemical property of polypeptide

Example 4-2(a)

Biological activity

Blood were collected from a healthy donor by using a syringe containing heparin, and then diluted with 2-fold volume of serum-free RPMI-1640 medium (pH 7.4). The blood was overlaid on ficoll, commercialized by Pharmacia LKB Biotechnology AB, Upsalla, Sweden, and centrifuged to obtain lymphocytes, which were then washed with RPMI-

1640 medium containing 10 v/v % bovine fetal serum before being suspended in a fresh preparation of the same medium to give a cell density of 5 x 10^5 cells/ml. 0.15 ml aliquots of the cell suspension was distributed into wells of micro plates with 96 wells.

To the wells with the cells were distributed 0.05 ml aliquots of solutions of the polypeptide in Example 4-1, diluted with RPMI-1640 medium (pH 7.4) containing 10 v/v % bovine fetal serum to give desired concentrations. 0.05 ml aliquots of fresh preparations of the same medium with 2.5 µg/ml concanavalin A were further added to the wells, before culturing in a 5 v/v % CO_2 incubator at 37°C for 24 hr. After the cultivation, 0.1 ml of the culture supernatant was collected from each well and examined on IFN-γ by usual enzyme immunoassay. In parallel, a systems as a control using the polypeptide in Reference for that in Example 4-1 or using no polypeptide was treated similarly as above. The results were in Table 1. IFN-γ in Table 1 were expressed with international units (IU), calculated based on the IFN-γ standard, Gg23-901-530, obtained from the International Institute of Health, USA

Table 1

Sample of polypeptide	IFN-γ production (IU/ml)
Example 4-2(a)	3.4 x 10^5
Example for Reference	1.7 x 10^5

Table 1 indicates that the lymphocytes as immunocompetent cells produce IFN-γ by the action of the present polypeptide.

It is more remarkable that the polypeptide in Example 4-1 could induce IFN-γ production more than that in Example for Reference. Considering this and the difference in the yields of the polypeptides, described in Example for Reference, it can be presumed: Even if DNAs could be substantially equivalent in encoding the same amino acid sequence, not only the expressing efficiencies of the DNAs may differ, but the products expressed by them may significantly differ in their biological activities as a result of post-translational modifications by intracellular enzymes, depending on types of the DNAs and their hosts; (a) one type is used a transformant formed by introducing a DNA, which is a cDNA, into a microorganisms as a host, and (b) other type is used a transformant formed by introducing the present genomic DNA into a mammalian cell as a host.

Example 4-2(b)

Molecular weight

SDS-polyacrylamide gel electrophoresis of the polypeptide in Example 4-1 in the presence of 2 w/v % dithiothreitol as a reducing agent, according to the method reported by U. K. Laemli et al., in "Nature", Vol.227, pp.680-685 (1970), exhibited a main band of a protein capable of inducing IFN-γ in a position corresponding to a molecular weight of about 18,000-19,500 daltons. The molecular weight makers used in the analysis were bovine serum albumin (67,000 daltons), ovalbumin (45,000 daltons), carbonic anhydrase (30,000 daltons), soy bean trypsin inhibitor (20,100 daltons) and α-lactoalbumin (14,000 daltons).

Example 4-2(c)

N-Terminal amino acid sequence

Conventional analysis using "MODEL 473A", a protein sequencer commercialized by Perkin-Elmer Corp., Norwalk, USA, revealed that the polypeptide in Example 4-1 had the amino acid sequence of SEQ ID NO:15 in the N-terminal region.

Judging collectively from this result as well as the information that SDS-polyacrylamide gel electrophresis exhibited a main band in a position corresponding to a molecular weight of about 18,000-19,500 daltons, and that the molecular weight calculated from the amino acid sequence of SEQ ID NO:1 was 18,199 daltons, it can be concluded that the polypeptide in Example 4-1 has the amino acid sequence of SEQ ID NO:6.

As is described above, the present invention is made based on the identification of a genomic DNA encoding the polypeptide which induces the production of IFN-γ by immunocompetent cells. The present genomic DNA efficiently express the present polypeptide when introduced into mammalian host cells. The polypeptide features higher biological activities than that obtained by the cDNA expression in *Escherichia coli*. Therefore, the present genomic DNA is useful for the recombinant DNA techniques to prepare the polypeptide capable of inducing IFN-γ production by immunocompetent cells. The present genomic DNA is useful to gene therapy for diseases including viral diseases, bacterial-infectious diseases, malignant tumors and immunopathies.

Thus, the present invention is a significant invention which has a remarkable effect and gives a great contribution to this field.

While there has been described what is at present considered to be the preferred embodiments of the present invention, it will be understood the various modifications may be made therein, and it is intended to cover in the appended claims all such modifications as fall within the true spirits and scope of the invention.

SEQUENCE LISTING

(1) GENERAL INFORMATION:

(i) APPLICANT:
NAME:KABUSHIKI KAISHA HAYASHIBARA SEIBUTSU KAGAKU KENKYUJO

(ii) TITLE OF INVENTION:GENOMIC DNA ENCODING A POLYPEPTIDE CAPABLE OF INDUCING THE PRODUCTION OF INTERFERON-γ

(iii) NUMBER OF SEQUENCES:15

(iv) ADDRESS:
(A) ADDRESSEE:KABUSHIKI KAISHA HAYASHIBARA SEIBUTSU KAGAKU KENKYUJO
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(E) COUNTRY:JAPAN
(F) POSTAL CODE (ZIP):700

(v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE:Floppy disk
(B) COMPUTER:IBM PC compatible
(C) OPERATING SYSTEM:PC-DOS/MS-DOS

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER:JP 185,305/96
(B) FILING DATE:June 27, 1996

(2) INFORMATION FOR SEQ ID NO:1:
(i)SEQUENCE CHARACTERISTICS:
(A)LENGTH:157 amino acids
(B)TYPE:amino acid
(D)TOPOLOGY:linear
(ii)MOLECULE TYPE:peptide
(xi)SEQUENCE DESCRIPTION:SEQ ID NO:1:

```
Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser Val Ile Arg Asn Leu Asn
1               5                  10                  15
Asp Gln Val Leu Phe Ile Asp Gln Gly Asn Arg Pro Leu Phe Glu Asp
            20                  25                  30
Met Thr Asp Ser Asp Cys Arg Asp Asn Ala Pro Arg Thr Ile Phe Ile
        35                  40                  45
Ile Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met Ala Val Thr Ile
    50                  55                  60
Ser Val Lys Cys Glu Lys Ile Ser Xaa Leu Ser Cys Glu Asn Lys Ile
65                  70                  75                  80
Ile Ser Phe Lys Glu Met Asn Pro Pro Asp Asn Ile Lys Asp Thr Lys
                85                  90                  95
Ser Asp Ile Ile Phe Phe Gln Arg Ser Val Pro Gly His Asp Asn Lys
            100                 105                 110
Met Gln Phe Glu Ser Ser Ser Tyr Glu Gly Tyr Phe Leu Ala Cys Glu
        115                 120                 125
Lys Glu Arg Asp Leu Phe Lys Leu Ile Leu Lys Lys Glu Asp Glu Leu
    130                 135                 140
Gly Asp Arg Ser Ile Met Phe Thr Val Gln Asn Glu Asp
145                 150                 155
```

(3) INFORMATION FOR SEQ ID NO:2:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH:1120 base pairs
(B) TYPE:nucleic acid
(C) STRANDEDNESS:double
(D) TOPOLOGY:linear
(ii) MOLECULE TYPE:cDNA to mRNA
(iii) HYPOTHETICAL:No
(iv) ANTI-SENSE:No
(vi) ORIGINAL SOURCE:
(A) ORGANISM:human
(F) TISSUE TYPE:liver
(iX) FEATURE:
(A) NAME/KEY:5'UTR
(B) LOCATION:1..177
(C) IDENTIFICATION METHODS:E
(A) NAME/KEY:leader peptide
(B) LOCATION:178..285
(C) IDENTIFICATION METHODS:S
(A) NAME/KEY:mat peptide
(B) LOCATION:286..756
(C) IDENTIFICATION METHODS:S
(A) NAME/KEY:3'UTR
(B) LOCATION:757..1120
(C) IDENTIFICATION METHODS:E
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

```
GCCTGGACAG TCAGCAAGGA ATTGTCTCCC AGTGCATTTT GCCCTCCTGG CTGCCAACTC   60
TGGCTGCTAA AGCGGCTGCC ACCTGCTGCA GTCTACACAG CTTCGGGAAG AGGAAAGGAA  120
CCTCAGACCT TCCAGATCGC TTCCTCTCGC AACAAACTAT TTGTCGCAGG AATAAAG     177
ATG GCT GCT GAA CCA GTA GAA GAC AAT TGC ATC AAC TTT GTG GCA ATG     225
Met Ala Ala Glu Pro Val Glu Asp Asn Cys Ile Asn Phe Val Ala Met
    -35                 -30                 -25
AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA GCT GAA GAT GAT GAA AAC     273
Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala Glu Asp Asp Glu Asn
-20                 -15                 -10                 -5
CTG GAA TCA GAT TAC TTT GGC AAG CTT GAA TCT AAA TTA TCA GTC ATA     321
Leu Glu Ser Asp Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser Val Ile
                1               5                  10
AGA AAT TTG AAT GAC CAA GTT CTC TTC ATT GAC CAA GGA AAT CGG CCT     369
Arg Asn Leu Asn Asp Gln Val Leu Phe Ile Asp Gln Gly Asn Arg Pro
        15                  20                  25
CTA TTT GAA GAT ATG ACT GAT TCT GAC TGT AGA GAT AAT GCA CCC CGG     417
Leu Phe Glu Asp Met Thr Asp Ser Asp Cys Arg Asp Asn Ala Pro Arg
    30                  35                  40
ACC ATA TTT ATT ATA AGT ATG TAT AAA GAT AGC CAG CCT AGA GGT ATG     465
Thr Ile Phe Ile Ile Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met
45                  50                  55                  60
GCT GTA ACT ATC TCT GTG AAG TGT GAG AAA ATT TCA AYT CTC TCC TGT     513
Ala Val Thr Ile Ser Val Lys Cys Glu Lys Ile Ser Xaa Leu Ser Cys
                65                  70                  75
GAG AAC AAA ATT ATT TCC TTT AAG GAA ATG AAT CCT CCT GAT AAC ATC     561
Glu Asn Lys Ile Ile Ser Phe Lys Glu Met Asn Pro Pro Asp Asn Ile
            80                  85                  90
AAG GAT ACA AAA AGT GAC ATC ATA TTC TTT CAG AGA AGT GTC CCA GGA     609
Lys Asp Thr Lys Ser Asp Ile Ile Phe Phe Gln Arg Ser Val Pro Gly
        95                  100                 105
CAT GAT AAT AAG ATG CAA TTT GAA TCT TCA TCA TAC GAA GGA TAC TTT     657
His Asp Asn Lys Met Gln Phe Glu Ser Ser Ser Tyr Glu Gly Tyr Phe
    110                 115                 120
CTA GCT TGT GAA AAA GAG AGA GAC CTT TTT AAA CTC ATT TTG AAA AAA     705
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Leu Ala Cys Glu Lys Glu Arg Asp Leu Phe Lys Leu Ile Leu Lys Lys
125                 130                 135                 140
GAG GAT GAA TTG GGG GAT AGA TCT ATA ATG TTC ACT GTT CAA AAC GAA    753
Glu Asp Glu Leu Gly Asp Arg Ser Ile Met Phe Thr Val Gln Asn Glu
                145                 150                 155
GAC TAGCTATTAA AATTTCATGC CGGGCGCAGT GGCTCACGCC TGTAATCCCA          806
Asp
GCCCTTTGGG AGGCTGAGGC GGGCAGATCA CCAGAGGTCA GGTGTTCAAG ACCAGCCTGA  866
CCAACATGGT GAAACCTCAT CTCTACTAAA AATACTAAAA ATTAGCTGAG TGTAGTGACG  926
CATGCCCTCA ATCCCAGCTA CTCAAGAGGC TGAGGCAGGA GAATCACTTG CACTCCGGAG  986
GTAGAGGTTG TGGTGAGCCG AGATTGCACC ATTGCGCTCT AGCCTGGGCA ACAACAGCAA 1046
AACTCCATCT CAAAAAATAA AATAAATAAA TAAACAAATA AAAAATTCAT AATGTGAAAA 1106
AAAAAAAAAA AAAA                                                   1120
```

(4) INFORMATION FOR SEQ ID NO:3:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH:135 base pairs
- (B) TYPE:nucleic acid
- (C) STRANDEDNESS:double
- (D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:
- (A) ORGANISM:human
- (F) TISSUE TYPE:placenta

(iX) FEATURE:
- (A) NAME/KEY:exon
- (B) LOCATION:1..135
- (C) IDENTIFICATION METHODS:S

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:3:

```
 AA AAC CTG GAA TCA GAT TAC TTT GGC AAG CTT GAA TCT AAA TTA TCA      47
Glu Asn Leu Glu Ser Asp Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser
  5                   1                   5                  10
GTC ATA AGA AAT TTG AAT GAC CAA GTT CTC TTC ATT GAC CAA GGA AAT      95
Val Ile Arg Asn Leu Asn Asp Gln Val Leu Phe Ile Asp Gln Gly Asn
                15                  20                  25
CGG CCT CTA TTT GAA GAT ATG ACT GAT TCT GAC TGT AGA G               135
Arg Pro Leu Phe Glu Asp Met Thr Asp Ser Asp Cys Arg Asp
            30                  35                  40
```

(5) INFORMATION FOR SEQ ID NO:4:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH:134 base pairs
- (B) TYPE:nucleic acid
- (C) STRANDEDNESS:double
- (D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:
- (A) ORGANISM:human
- (F) TISSUE TYPE:placenta

(iX) FEATURE:
- (A) NAME/KEY:exon
- (B) LOCATION:1..134
- (C) IDENTIFICATION METHODS:S

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:

```
 AT AAT GCA CCC CGG ACC ATA TTT ATT ATA AGT ATG TAT AAA GAT AGC      47
Asp Asn Ala Pro Arg Thr Ile Phe Ile Ile Ser Met Tyr Lys Asp Ser
40                  45                  50                  55
```

```
CAG CCT AGA GGT ATG GCT GTA ACT ATC TCT GTG AAG TGT GAG AAA ATT      95
Gln Pro Arg Gly Met Ala Val Thr Ile Ser Val Lys Cys Glu Lys Ile
                60                  65                  70
TCA ACT CTC TCC TGT GAG AAC AAA ATT ATT TCC TTT AAG                 134
Ser Thr Leu Ser Cys Glu Asn Lys Ile Ile Ser Phe Lys
            80                  85
```

(6) INFORMATION FOR SEQ ID NO:5:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH:87 base pairs
- (B) TYPE:nucleic acid
- (C) STRANDEDNESS:double
- (D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:
- (A) ORGANISM:human
- (F) TISSUE TYPE:placenta

(iX) FEATURE:
- (A) NAME/KEY:exon
- (B) LOCATION:1..87
- (C) IDENTIFICATION METHODS:S

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:

```
GAATAAAG ATG GCT GCT GAA CCA GTA GAA GAC AAT TGC ATC AAC TTT GTG     50
         Met Ala Ala Glu Pro Val Glu Asp Asn Cys Ile Asn Phe Val
            -35                 -30                 -25
GCA ATG AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA G                   87
Ala Met Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala
        -20                 -15                 -10
```

(7) INFORMATION FOR SEQ ID NO:6:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH:12 base pairs
- (B) TYPE:nucleic acid
- (C) STRANDEDNESS:double
- (D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:
- (A) ORGANISM:human
- (F) TISSUE TYPE:placenta

(iX) FEATURE:
- (A) NAME/KEY:exon
- (B) LOCATION:1..87
- (C) IDENTIFICATION METHODS:S

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:6:

```
 CT GAA GAT GAT G                                                   12
Ala Glu Asp Asp Glu
-10
```

(8) INFORMATION FOR SEQ ID NO:7:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH:2167 base pairs
- (B) TYPE:nucleic acid
- (C) STRANDEDNESS:double
- (D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:

(A) ORGANISM: human
(F) TISSUE TYPE: placenta
(iX) FEATURE:
(A) NAME/KEY: exon + 3'UTR
(B) LOCATION: 1..2167
(C) IDENTIFICATION METHODS: E
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:7:

```
GAA ATG AAT CCT CCT GAT AAC ATC AAG GAT ACA AAA AGT GAC ATC ATA      48
Glu Met Asn Pro Pro Asp Asn Ile Lys Asp Thr Lys Ser Asp Ile Ile
85                  90                  95                  100
TTC TTT CAG AGA AGT GTC CCA GGA CAT GAT AAT AAG ATG CAA TTT GAA      96
Phe Phe Gln Arg Ser Val Pro Gly His Asp Asn Lys Met Gln Phe Glu
                105                 110                 115
TCT TCA TCA TAC GAA GGA TAC TTT CTA GCT TGT GAA AAA GAG AGA GAC     144
Ser Ser Ser Tyr Glu Gly Tyr Phe Leu Ala Cys Glu Lys Glu Arg Asp
            120                 125                 130
CTT TTT AAA CTC ATT TTG AAA AAA GAG GAT GAA TTG GGG GAT AGA TCT     192
Leu Phe Lys Leu Ile Leu Lys Lys Glu Asp Glu Leu Gly Asp Arg Ser
        135                 140                 145
ATA ATG TTC ACT GTT CAA AAC GAA GAC TAGCTAT TAAAATTTCA TGCCGGGCGC   246
Ile Met Phe Thr Val Gln Asn Glu Asp
    150                 155
AGTGGCTCAC GCCTGTAATC CCAGCCCTTT GGGAGGCTGA GGCGGGCAGA TCACCAGAGG   306
TCAGGTGTTC AAGACCAGCC TGACCAACAT GGTGAAACCT CATCTCTACT AAAAATACAA   366
AAAATTAGCT GAGTGTAGTG ACCCATGCCC TCAATCCCAG CTACTCAAGA GGCTGAGGCA   426
GGAGAATCAC TTGCACTCCG GAGGTGGAGG TTGTGGTGAG CCGAGATTGC ACCATTGCGC   486
TCTAGCCTGG GCAACAACAG CAAAACTCCA TCTCAAAAAA TAAAATAAAT AAATAAACAA   546
ATAAAAAATT CATAATGTGA ACTGTCTGAA TTTTTATGTT TAGAAAGATT ATGAGATTAT   606
TAGTCTATAA TTGTAATGGT GAAATAAAAT AAATACCAGT CTTGAAAAAC ATCATTAAGA   666
AATGAATGAA CTTTCACAAA AGCAAACAAA CAGACTTTCC CTTATTTAAG TGAATAAAAT   726
AAAATAAAAT AAAATAATGT TTAAAAAATT CATAGTTTGA AAACATTCTA CATTGTTAAT   786
TGGCATATTA ATTATACTTA ATATAATTAT TTTTAAATCT TTTGGGTTAT TAGTCCTAAT   846
GACAAAAGAT ATTGATATTT GAACTTTCTA ATTTTTAAGA ATATCGTTAA ACCATCAATA   906
TTTTTATAAG GAGGCCACTT CACTTGACAA ATTTCTGAAT TTCCTCCAAA GTCAGTATAT   966
TTTTAAAATT CAGTTTGATC CTGAATCCAG CAATATATAA AAGGGATTAT ATACTCTGGC  1026
CAACTGACAT TCATCCTAGG AATGCAAAGA TGGTTTAATA TCCTAAAATC AATTAACATA  1086
ACATACTATA TTAATAAAGT ATCAAAACAG TATTCTCATC TTTTTTTCTT TTTTCACAAT  1146
TCCTTGGTTA CACTATCATC TCAATAGATG CAGAAAAAGC ATTTGACAAA ATCCAATTCA  1206
TAATAAAAAT TCTCAAACTT GAAAGAGAAC ATCATAAAGG CATCTATGAA AAACCTACAG  1266
CTAATATCAT ACTTAACGAT GAAAAACTGA ATTATTTTAC CCTAAGATCA AGAATAATGC  1326
AAGCATGTCA GCTCTTGCAA CTTCTATTCA ACATTGTACT GGAGGTTCTA GCCAGAGCAA  1386
CCATACAATA AATAAAAATA AAAGGCACCC AGATTAGAAA GGAAGTCTTT ATTTGCAGAC  1446
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TCAGCAAGGT TGCAGGTTGC AATATCAATA TGCAAAAATA CATTGAAGGC TGGGCTCAGT  1566
GGAGATGGCA TGTACCTTTC GTCCCAGCTA CTTGGGAGGC TGAGGTAGGA GGATCACTTG  1626
AGGTGAGGAG TTTGAGGCTA TAGTGCAATG TGATCTTGCC TGTGAATAGC CACTGCACTC  1686
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T                                                                  2167
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(9) INFORMATION FOR SEQ ID NO:8:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 1334 base pairs

(B)TYPE:nucleic acid
(C)STRANDEDNESS:double
(D)TOPOLOGY:linear
(ii)MOLECULE TYPE:Genomic DNA
(vi)ORIGINAL SOURCE:
(A)ORGANISM:human
(F)TISSUE TYPE:placenta
(iX)FEATURE:
(A)NAME/KEY:intron
(B)LOCATION:1..1334
(C)IDENTIFICATION METHODS:E
(xi)SEQUENCE DESCRIPTION: SEQ ID NO:8:

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(10) INFORMATION FOR SEQ ID NO:9:
(i)SEQUENCE CHARACTERISTICS:
(A)LENGTH:4773 base pairs
(B)TYPE:nucleic acid
(C)STRANDEDNESS:double
(D)TOPOLOGY:linear
(ii)MOLECULE TYPE:Genomic DNA
(vi)ORIGINAL SOURCE:
(A)ORGANISM:human
(F)TISSUE TYPE:placenta
(iX)FEATURE:
(A)NAME/KEY:intron
(B)LOCATION:1..4773
(C)IDENTIFICATION METHODS:E
(xi)SEQUENCE DESCRIPTION: SEQ ID NO:9:

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(11) INFORMATION FOR SEQ ID NO:10:

(i) SEQUENCE CHARACTERISTICS:
- (A) LENGTH: 8835 base pairs
- (B) TYPE: nucleic acid
- (C) STRANDEDNESS: double
- (D) TOPOLOGY: linear

(ii) MOLECULE TYPE: Genomic DNA

(vi) ORIGINAL SOURCE:
- (A) ORGANISM: human
- (F) TISSUE TYPE: placenta

(iX) FEATURE:
- (A) NAME/KEY: intron
- (B) LOCATION: 1..8835
- (C) IDENTIFICATION METHODS: E

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:10:

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CACATCACAT GTTAGCATAA GTAACATTTT TAATTAAAAA TAACTATGTA CTTTTTTAAC  300
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GAGCTCACAG CCTAGTCATT ATTGTTCTGA TTTTTAATAT TAATGTTGGT TTGGGTTTTG 6900
GTGAAAAATG TTTAGACTTA TCTTAGTGAT CTTTTCATCC TTTGCTATAT TATTTTTCTC 6960
TAAGAGTCTT CCTTATCCCC TCCTTTAAAA AACTAGGTGA TAATTCTAAA TTGTAAATTT 7020
AAATATTATA AATAGCTTAT AAAATTTAAT ATTTATAATA TTTAAATGTT TGATAAATAT 7080
TTAAATTTTA TAATATTTAA ATGTTTATTT AAATTCATTT GTACATCAGT TTTTATTTTA 7140
TTTAAATGTG TTGGCCAGGC ATGGTGGCTG ACACCTATAA TCCCAGAACT TTGAGAGGCC 7200
AAGTCAGGCA AACCATTTGA GCTCAGGAGT TTGAGACCAC CCTGGGCAAC GTGGTGAAAC 7260
CCTGTCTCTA CCAAACATAT GAAAACTTAT CTGGGTGTGG TGGCACGCAT CTGTGGTCCC 7320
AGATGGGAGT CCCAGGCTAA GATGGGAGAA TCGCTTGAAC CCAGGTGAGA GGGGTGGGGT 7380
GGATGTTGCA GTGAGCTGAG ATCGTGCCAC TGCACTCCAA CCTGGGTGAC AGAGTGAGAC 7440
TCCATCTCAA AAAAAAAAAA TGTTATCTAA ATAAGATAAA TTTAATAACT GTTCGCACTT 7500
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AGCACAGATT TATGAATACT CTCCTGCATA CCATTTAATT TTTAAACAAA TTTTAATGCA 7800
GTATATATGT GCCTTTTTAC CAACACATTA AATAATAAGA TCTACTGTGA GGACTAAATT 7860
TCTGTAATTT CAAAGTAGTA ATGAGTTTAA ACCATGTCTC AAGATCTCTG CAATAACTGT 7920
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TGTGGTTTGT TTCCTGCAAC TAATCATGGG AGGAATGCTA AATTTCAGAG GTTGGTGAAA 8040
ATACATGTGT ATTTTTTTCC CCATCCAAGT TCACAGATTT CTCACACTGA GAACTCCTAT 8100
TCCATAACAA AATTCTGGAA GCCTGCACAC CGTATTGGAA GAAGGGCAGA AAGGAAAAGC 8160
AAATGGAAGG ATTTAAATTT TTTTCAAATC CTGTATCCCT TGATTTTACA GCAAGATTGT 8220
ATTTATGTAT TACTTGTGTT AAAAATATAG TATAATCGAG ACTCCAGATC AAAAATCACC 8280
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CCTGTACCCT CAGATGGAAG CACTTTTTTA TCATTGTTTC ACCTTTAGCA TTTTGACAAT 8400
GAAGTCACAA ACCTTCAGCC TCTCACCCAT AGGAACCCAC TGGTTGTAAG AGAAGGATGA 8460
AGCCAGTCCT TCCTAAAGGG CACGATTAGA TGTGTTTATG GCATCCTCAG GTGAAACTAT 8520
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TACTAGGCCA TTTATCTACC CTTTATAATA TTGTTTAATG AGAAAATGCT TTCTATCTTC 8640
CAAATATCTG ATGATTTGTA AGAGAACACT TAAACATGGG TATTCATAAG CTGAAACTTC 8700
TGGCATTTAT TGAATGTCAA GATTGTTCAT CAGTATACTA GGTGATTAAC TGACCACTGA 8760
ACTTGAAGGT AGTATAAAGT AGTAGTAAAA GGTACAATCA TTGTCTCTTA ACAGATGGCT 8820
CTTTGCTTTC ATTAG                                                  8835
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(12) INFORMATION FOR SEQ ID NO:11:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH:1371 base pairs

(B) TYPE:nucleic acid
(C) STRANDEDNESS:double
(D) TOPOLOGY:linear
(ii) MOLECULE TYPE:Genomic DNA
(vi) ORIGINAL SOURCE:
(A) ORGANISM:human
(F) TISSUE TYPE:placenta
(iX) FEATURE:
(A) NAME/KEY:intron
(B) LOCATION:1..1371
(C) IDENTIFICATION METHODS:E
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

GTAAGGCTAA TGCCATAGAA CAAATACCAG GTTCAGATAA ATCTATTCAA TTAGAAAAGA 60
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GGCTCTTAAA AAAATAGTGG ACCTCTAGAA ATTAACCACA ACATGTCCAA GGTCTCAGCA 180
CCTTGTCACA CCACGTGTCC TGGCACTTTA ATCAGCAGTA GCTCACTCTC CAGTTGGCAG 240
TAAGTGCACA TCATGAAAAT CCCAGTTTTC ATGGGAAAAT CCCAGTTTTC ATTGGATTTC 300
CATGGGAAAA ATCCCAGTAC AAAACTGGGT GCATTCAGGA AATACAATTT CCCAAAGCAA 360
ATTGGCAAAT TATGTAAGAG ATTCTCTAAA TTTAGAGTTC CGTGAATTAC ACCATTTTAT 420
GTAAATATGT TTGACAAGTA AAAATTGATT CTTTTTTTTT TTTTCTGTTG CCCAGGCTGG 480
AGTGCAGTGG CACAATCTCT GCTCACTGCA ACCTCCACCT CCTGGGTTCA AGCAATTCTC 540
CTGCCTCAGC CTTCTGAGTA GCTGGGACTA CAGGTGCATC CCGCCATGCC TGGCTAATTT 600
TTGGGTATTT TTACTAGAGA CAGGGTTTTG GCATGTTGTC CAGGCTGGTC TTGGACTCCT 660
GATCTCAGAT GATCCTCCTG GCTCGGGCTC CCAAAGTGCT GGGATTACAG GCATGAACCA 720
CCACACATGG CCTAAAAATT GATTCTTATG ATTAATCTCC TGTGAACAAT TTGGCTTCAT 780
TTGAAAGTTT GCCTTCATTT GAAACCTTCA TTTAAAAGCC TGAGCAACAA AGTGAGACCC 840
CATCTCTACA AAAAACTGCA AAATATCCTG TGGACACCTC CTACCTTCTG TGGAGGCTGA 900
AGCAGGAGGA TCACTTGAGC CTAGGAATTT GAGCCTGCAG TGAGCTATGA TCCCACCCCT 960
ACACTCCAGC CTGCATGACA GTAGACCCTG ACACACACAC ACAAAAAAAA ACCTTCATAA 1020
AAAATTATTA GTTGACTTTT CTTAGGTGAC TTTCCGTTTA AGCAATAAAT TTAAAAGTAA 1080
AATCTCTAAT TTTAGAAAAT TTATTTTTAG TTACATATTG AAATTTTTAA ACCCTAGGTT 1140
TAAGTTTTAT GTCTAAATTA CCTGAGAACA CACTAAGTCT GATAAGCTTC ATTTTATGGG 1200
CCTTTTGGAT GATTATATAA TATTCTGATG AAAGCCAAGA CAGACCCTTA AACCATAAAA 1260
ATAGGAGTTC GAGAAAGAGG AGTAGCAAAA GTAAAAGCTA GAATGAGATT GAATTCTGAG 1320
TCGAAATACA AAATTTTACA TATTCTGTTT CTCTCTTTTT CCCCCTCTTA G 1371

(13) INFORMATION FOR SEQ ID NO:12:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH:3383 base pairs
(B) TYPE:nucleic acid
(C) STRANDEDNESS:double
(D) TOPOLOGY:linear
(ii) MOLECULE TYPE:Genomic DNA
(vi) ORIGINAL SOURCE:
(A) ORGANISM:human
(F) TISSUE TYPE:placenta
(iX) FEATURE:
(A) NAME/KEY:intron
(B) LOCATION:1..3383
(C) IDENTIFICATION METHODS:E
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:12:

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TCTCACCATT GTCAGCTGAG GAAAAAAAAA AATGGTTCTC ATGCTACCAA TCTGCCTTCA 120
AAGAAATGTG GACTCAGTAG CACAGCTTTG GAATGAAGAT GATCATAAGA GATACAAAGA 180
AGAACCTCTA GCAAAAGATG CTTCTCTATG CCTTAAAAAA TTCTCCAGCT CTTAGAATCT 240
ACAAAATAGA CTTTGCCTGT TTCATTGGTC CTAAGATTAG CATGAAGCCA TGGATTCTGT 300
TGTAGGGGGA GCGTTGCATA GGAAAAAGGG ATTGAAGCAT TAGAATTGTC CAAAATCAGT 360
AACACCTCCT CTCAGAAATG CTTTGGGAAG AAGCCTGGAA GGTTCCGGGT TGGTGGTGGG 420

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GTGGGGCAGA AAATTCTGGA AGTAGAGGAG ATAGGAATGG GTGGGGCAAG AAGACCACAT   480
TCAGAGGCCA AAAGCTGAAA GAAACCATGG CATTTATGAT GAATTCAGGG TAATTCAGAA   540
TGGAAGTAGA GTAGGAGTAG GAGACTGGTG AGAGGAGCTA GAGTGATAAA CAGGGTGTAG   600
AGCAAGACGT TCTCTCACCC CAAGATGTGA AATTTGGACT TTATCTTGGA GATAATAGGG   660
TTAATTAAGC ACAATATGTA TTAGCTAGGG TAAAGATTAG TTTGTTGTAA CAAAGACATC   720
CAAAGATACA GTAGCTGAAT AAGATAGAGA ATTTTTCTCT CAAAGAAAGT CTAAGTAGGC   780
AGCTCAGAAG TAGTATGGCT GGAAGCAACC TGATGATATT GGGACCCCCA ACCTTCTTCA   840
GTCTTGTACC CATCATCCCC TAGTTGTTGA TCTCACTCAC ATAGTTGAAA ATCATCATAC   900
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TAATCACATG GCCACACCAA GTGCAAGGAA ATCTGGAAAA TATAATCTTT ATTCCAGGTA  1080
GCCATATGAC TCTTTAAAAT TCAGAAATAA TATATTTTTA AAATATCATT CTGGCTTTGG  1140
TATAAAGAAT TGATGGTGTG GGGTGAGGAG GCCAAAATTA AGGGTTGAGA GCCTATTATT  1200
TTAGTTATTA CAAGAAATGA TGGTGTCATG AATTAAGGTA GACATAGGGG AGTGCTGATG  1260
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TGTACCATTC AGTGAAATAG GGAACACAGG AGGAAGAGCA GGTTTTGGTG TATACAAAGA  1440
GGAGGATGGA TGACGCATTT CGTTTTGGAT CTGAGATGTC TGTGGAACGT CCTAGTGGAG  1500
ATGTCCACAA ACTCTTCTAC ATGTGGTTCT GAGTTCAGGA CACAGATTTG GGCTGGAGAT  1560
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TACATTTAAA GGATGCAGTA GAAAGAAGCT AATAAACAAC AGAGAGCAGA CTAACCAAAA  1740
GGGGAGAAGA AAAACCAAGA GAATTCCACC GACTCCCAGG AGAGCATTTC AAGATTGAGG  1800
GGATAGGTGT TGTGTTGAAT TTTGCAGCCT TGAGAATCAA GGGCCAGAAC ACAGCTTTTA  1860
GATTTAGCAA CAAGGAGTTT GGTGATCTCA GTGAAAGCAG CTTGATGGTG AAATGGAGGC  1920
AGAGGCAGAT TGCAATGAGT GAAACAGTGA ATGGGAAGTG AAGAAATGAT ACAGATAATT  1980
CTTGCTAAAA GCTTGGCTGT TAAAAGGAGG AGAGAAACAA GACTAGCTGC AAAGTGAGAT  2040
TGGGTTGATG GAGCAGTTTT AAATCTCAAA ATAAAGAGCT TTGTGCTTTT TTGATTATGA  2100
AAATAATGTG TTAATTGTAA CTAATTGAGG CAATGAAAAA AGATAATAAT ATGAAAGATA  2160
AAAATATAAA AACCACCCAG AAATAATGAT AGCTACCATT TTGATACAAT ATTTCTACAC  2220
TCCTTTCTAT GTATATATAC AGACACAGAA ATGCTTATAT TTTTATTAAA AGGGATTGTA  2280
CTATACCTAA GCTGCTTTTT CTAGTTAGTG ATATATATGG ACATCTCTCC ATGGCAACGA  2340
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GAACGCTTGT GCCATTGTTC ACCAGCAGCA CTCAGCAGCC AAGTTTGGAG TTTTGTAGCA  2940
GAAAGACAAA TAAGTTAGGG ATTTAATATC CTGGCCAAAT GGTAGACAAA ATGAACTCTG  3000
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ATGAGATTTA ATGTTTATTG TAG                                          3383
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(14) INFORMATION FOR SEQ ID NO:13:

(i) SEQUENCE CHARACTERISTICS:

(A) LENGTH:11464 base pairs

(B) TYPE:nucleic acid

(C) STRANDEDNESS:double

(D) TOPOLOGY:linear

(ii) MOLECULE TYPE:Genomic DNA

(vi) ORIGINAL SOURCE:

(A) ORGANISM:human

(F)TISSUE TYPE:placenta
(iX)FEATURE:
(A)NAME/KEY:5'UTR
(B)LOCATION:1..3
(C)IDENTIFICATION METHODS:E
(A)NAME/KEY:leader peptide
(B)LOCATION:4..82
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:intron
(B)LOCATION:83..1453
(C)IDENTIFICATION METHODS:E
(A)NAME/KEY:leader peptide
(B)LOCATION:1454..1465
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:intron
(B)LOCATION:1466..4848
(C)IDENTIFICATION METHODS:E
(A)NAME/KEY:leader peptide
(B)LOCATION:4849..4865
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:mat peptide
(B)LOCATION:4866..4983
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:intron
(B)LOCATION:4984..6317
(C)IDENTIFICATION METHODS:E
(A)NAME/KEY:mat peptide
(B)LOCATION:6318..6451
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:intron
(B)LOCATION:6452..11224
(C)IDENTIFICATION METHODS:E
(A)NAME/KEY:mat peptide
(B)LOCATION:11225..11443
(C)IDENTIFICATION METHODS:S
(A)NAME/KEY:3'UTR
(B)LOCATION:11444..11464
(C)IDENTIFICATION METHODS:E
(xi)SEQUENCE DESCRIPTION: SEQ ID NO:13:

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ATG AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA G   GTAAGG CTAATGCCAT
98
Met Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala
    -20                 -15                 -10
AGAACAAATA CCAGGTTCAG ATAAATCTAT TCAATTAGAA AAGATGTTGT GAGGTGAACT
158
ATTAAGTGAC TCTTTGTGTC ACCAAATTTC ACTGTAATAT TAATGGCTCT TAAAAAAATA
218
GTGGACCTCT AGAAATTAAC CACAACATGT CCAAGGTCTC AGCACCTTGT CACACCACGT
278
GTCCTGGCAC TTTAATCAGC AGTAGCTCAC TCTCCAGTTG GCAGTAAGTG CACATCATGA
338
AAATCCCAGT TTTCATGGGA AAATCCCAGT TTTCATTGGA TTTCCATGGG AAAAATCCCA
398
GTACAAAACT GGGTGCATTC AGGAAATACA ATTTCCCAAA GCAAATTGGC AAATTATGTA
458
AGAGATTCTC TAAATTTAGA GTTCCGTGAA TTACACCATT TTATGTAAAT ATGTTTGACA
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518
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578
CTCTGCTCAC TGCAACCTCC ACCTCCTGGG TTCAAGCAAT TCTCCTGCCT CAGCCTTCTG
638
AGTAGCTGGG ACTACAGGTG CATCCCGCCA TGCCTGGCTA ATTTTTGGGT ATTTTTACTA
698
GAGACAGGGT TTTGGCATGT TGTCCAGGCT GGTCTTGGAC TCCTGATCTC AGATGATCCT
758
CCTGGCTCGG GCTCCCAAAG TGCTGGGATT ACAGGCATGA ACCACCACAC ATGGCCTAAA
818
AATTGATTCT TATGATTAAT CTCCTGTGAA CAATTTGGCT TCATTTGAAA GTTTGCCTTC
878
ATTTGAAACC TTCATTTAAA AGCCTGAGCA ACAAAGTGAG ACCCCATCTC TACAAAAAAC
938
TGCAAAATAT CCTGTGGACA CCTCCTACCT TCTGTGGAGG CTGAAGCAGG AGGATCACTT
998
GAGCCTAGGA ATTTGAGCCT GCAGTGAGCT ATGATCCCAC CCCTACACTC CAGCCTGCAT
1058
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1118
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1238
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1358
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Ala Glu Asp Asp Glu
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1950
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2010
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2070
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2190
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2250

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2430
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2550
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2610
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2670
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2850
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2970
CACAAACTCT TCTACATGTG GTTCTGAGTT CAGGACACAG ATTTGGGCTG GAGATAGAGA
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3570
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3870
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3990
CCCAGGAGGT AGAATTGCTG GGTTGATAGA AGAGAAAGGA TGATTGCCAA ATTAAAGCTT
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ATT GAC CAA GGA AAT CGG CCT CTA TTT GAA GAT ATG ACT GAT TCT GAC
4976
Ile Asp Gln Gly Asn Arg Pro Leu Phe Glu Asp Met Thr Asp Ser Asp
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Cys Arg Asp
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(15) INFORMATION FOR SEQ ID NO:14:

- (i) SEQUENCE CHARACTERISTICS:
  - (A) LENGTH:28994 base pairs
  - (B) TYPE:nucleic acid
  - (C) STRANDEDNESS:double
  - (D) TOPOLOGY:linear
- (ii) MOLECULE TYPE:Genomic DNA
- (vi) ORIGINAL SOURCE:
  - (A) ORGANISM:human
  - (F) TISSUE TYPE:placenta
- (iX) FEATURE:
  - (A) NAME/KEY:5'UTR
  - (B) LOCATION:1..15606
  - (C) IDENTIFICATION METHODS:E
  - (A) NAME/KEY:leader peptide
  - (B) LOCATION:15607..15685
  - (C) IDENTIFICATION METHODS:S
  - (A) NAME/KEY:intron
  - (B) LOCATION:15686..17056
  - (C) IDENTIFICATION METHODS:E
  - (A) NAME/KEY:leader peptide
  - (B) LOCATION:17057..17068
  - (C) IDENTIFICATION METHODS:S
  - (A) NAME/KEY:intron
  - (B) LOCATION:17069..20451

(C) IDENTIFICATION METHODS:E
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(C) IDENTIFICATION METHODS:S
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(C) IDENTIFICATION METHODS:E
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(B) LOCATION:21921..22054
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(C) IDENTIFICATION METHODS:E

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:14:

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GTAGTTCTGA ATGGTAACAA GAAATTGAGT GTGCCTTTGA GAGTAGGTTA AAAAACAATA
20315
GGCAACTTTA TTGTAGCTAC TTCTGGAACA GAAGATTGTC ATTAATAGTT TTAGAAAACT
20375
AAAATATATA GCATACTTAT TTGTCAATTA ACAAAGAAAC TATGTATTTT TAAATGAGAT
20435
TTAATGTTTA TTGTAG  AA AAC CTG GAA TCA GAT TAC TTT GGC AAG CTT GAA
20486
                  Glu Asn Leu Glu Ser Asp Tyr Phe Gly Lys Leu Glu
                     -5                  1                   5
TCT AAA TTA TCA GTC ATA AGA AAT TTG AAT GAC CAA GTT CTC TTC ATT
20534
Ser Lys Leu Ser Val Ile Arg Asn Leu Asn Asp Gln Val Leu Phe Ile
            10                  15                  20
GAC CAA GGA AAT CGG CCT CTA TTT GAA GAT ATG ACT GAT TCT GAC TGT
20582
Asp Gln Gly Asn Arg Pro Leu Phe Glu Asp Met Thr Asp Ser Asp Cys
        25                  30                  35
AGA G   GT ATTTTTTTTA ATTCGCAAAC ATAGAAATGA CTAGCTACTT CTTCCCATTC
20638
Arg Asp
    40
TGTTTTACTG CTTACATTGT TCCGTGCTAG TCCCAATCCT CAGATGAAAA GTCACAGGAG
20698
TGACAATAAT TTCACTTACA GGAAACTTTA TAAGGCATCC ACGTTTTTTA GTTGGGGTAA
20758
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AAAATTGGAT ACAATAAGAC ATTGCTAGGG GTCATGCCTC TCTGAGCCTG CCTTTGAATC
20818
ACCAATCCCT TTATTGTGAT TGCATTAACT GTTTAAAACC TCTATAGTTG GATGCTTAAT
20878
CCCTGCTTGT TACAGCTGAA AATGCTGATA GTTTACCAGG TGTGGTGGCA TCTATCTGTA
20938
ATCCTAGCTA CTTGGGAGGC TCAAGCAGGA GGATTGCTTG AGGCCAGGAC TTTGAGGCTG
20998
TAGTACACTG TGATCGTACC TGTGAATAGC CACTGCACTC CAGCCTGGGT GATATACAGA
21058
CCTTGTCTCT AAAATTAAAA AAAAAAAAAA AAAAAACCTT AGGAAAGGAA ATTGATCAAG
21118
TCTACTGTGC CTTCCAAAAC ATGAATTCCA AATATCAAAG TTAGGCTGAG TTGAAGCAGT
21178
GAATGTGCAT TCTTTAAAAA TACTGAATAC TTACCTTAAC ATATATTTTA AATATTTTAT
21238
TTAGCATTTA AAAGTTAAAA ACAATCTTTT AGAATTCATA TCTTTAAAAT ACTCAAAAAA
21298
GTTGCAGCGT GTGTGTTGTA ATACACATTA AACTGTGGGG TTGTTTGTTT GTTTGAGATG
21358
CAGTTTCACT CTGTCACCCA GGCTGAAGTG CAGTGCAGTG CAGTGGTGTG ATCTCGGCTC
21418
ACTACAACCT CCACCTCCCA CGTTCAAGCG ATTCTCATGC CTCAGTCTCC CGAGTAGGTG
21478
GGATTACAGG CATGCACCAC TTACACCCGG CTAATTTTTG TATTTTTAGT AGAGCTGGGG
21538
TTTCACCATG TTGGCCAGGC TGGTCTCAAA CCCCTAACCT CAAGTGATCT GCCTGCCTCA
21598
GCCTCCCAAA CAAACAAACA ACCCCACAGT TTAATATGTG TTACAACACA CATGCTGCAA
21658
CTTTTATGAG TATTTTAATG ATATAGATTA TAAAAGGTTG TTTTTAACTT TTAAATGCTG
21718
GGATTACAGG CATGAGCCAC TGTGCCAGGC CTGAACTGTG TTTTTAAAAA TGTCTGACCA
21778
GCTGTACATA GTCTCCTGCA GACTGGCCAA GTCTCAAAGT GGGAACAGGT GTATTAAGGA
21838
CTATCCTTTG GTTAAATTTC CGCAAATGTT CCTGTGCAAG AATTCTTCTA ACTAGAGTTC
21898
TCATTTATTA TATTTATTTC AG  AT AAT GCA CCC CGG ACC ATA TTT ATT ATA
21949
                          Asp Asn Ala Pro Arg Thr Ile Phe Ile Ile
                          40                  45
AGT ATG TAT AAA GAT AGC CAG CCT AGA GGT ATG GCT GTA ACT ATC TCT
21997
Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met Ala Val Thr Ile Ser
50                  55                  60                  65
GTG AAG TGT GAG AAA ATT TCA ACT CTC TCC TGT GAG AAC AAA ATT ATT
22045
Val Lys Cys Glu Lys Ile Ser Thr Leu Ser Cys Glu Asn Lys Ile Ile
                70                  75                  80
TCC TTT AAG GTAAGACTG AGCCTTACTT TGTTTTCAAT CATGTTAATA TAATCAATAT
22103
Ser Phe Lys
AATTAGAAAT ATAACATTAT TTCTAATGTT AATATAAGTA ATGTAATTAG AAAACTCAAA
22163
TATCCTCAGA CCAACCTTTT GTCTAGAACA GAAATAACAA GAAGCAGAGA ACCATTAAAG
22223
TGAATACTTA CTAAAAATTA TCAAACTCTT TACCTATTGT GATAATGATG GTTTTTCTGA
22283
GCCTGTCACA GGGGAAGAGG AGATACAACA CTTGTTTTAT GACCTGCATC TCCTGAACAA
22323
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TCAGTCTTTA TACAAATAAT AATGTAGAAT ACATATGTGA GTTATACATT TAAGAATAAC
22403
ATGTGACTTT CCAGAATGAG TTCTGCTATG AAGAATGAAG CTAATTATCC TTCTATATTT
22463
CTACACCTTT GTAAATTATG ATAATATTTT AATCCCTAGT TGTTTTGTTG CTGATCCTTA
22523
GCCTAAGTCT TAGACACAAG CTTCAGCTTC CAGTTGATGT ATGTTATTTT TAATGTTAAT
22583
CTAATTGAAT AAAAGTTATG AGATCAGCTG TAAAAGTAAT GCTATAATTA TCTTCAAGCC
22643
AGGTATAAAG TATTTCTGGC CTCTACTTTT TCTCTATTAT TCTCCATTAT TATTCTCTAT
22703
TATTTTTCTC TATTTCCTCC ATTATTGTTA GATAAACCAC AATTAACTAT AGCTACAGAC
22763
TGAGCCAGTA AGAGTAGCCA GGGATGCTTA CAAATTGGCA ATGCTTCAGA GGAGAATTCC
22823
ATGTCATGAA GACTCTTTTT GAGTGGAGAT TTGCCAATAA ATATCCGCTT TCATGCCCAC
22883
CCAGTCCCCA CTGAAAGACA GTTAGGATAT GACCTTAGTG AAGGTACCAA GGGGCAACTT
22943
GGTAGGGAGA AAAAAGCCAC TCTAAAATAT AATCCAAGTA AGAACAGTGC ATATGCAACA
23003
GATACAGCCC CCAGACAAAT CCCTCAGCTA TCTCCCTCCA ACCAGAGTGC CACCCCTTCA
23063
GGTGACAATT TGGAGTCCCC ATTCTAGACC TGACAGGCAG CTTAGTTATC AAAATAGCAT
23123
AAGAGGCCTG GGATGGAAGG GTAGGGTGGA AAGGGTTAAG CATGCTGTTA CTGAACAACA
23183
TAATTAGAAG GGAAGGAGAT GGCCAAGCTC AAGCTATGTG GGATAGAGGA AAACTCAGCT
23243
GCAGAGGCAG ATTCAGAAAC TGGGATAAGT CCGAACCTAC AGGTGGATTC TTGTTGAGGG
23303
AGACTGGTGA AAATGTTAAG AAGATGGAAA TAATGCTTGG CACTTAGTAG GAACTGGGCA
23363
AATCCATATT TGGGGGAGCC TGAAGTTTAT TCAATTTTGA TGGCCCTTTT AAATAAAAAG
23423
AATGTGGCTG GGCGTGGTGG CTCACACCTG TAATCCCAGC ACTTTGGGAG GCCGAGGGGG
23483
GCGGATCACC TGAAGTCAGG AGTTCAAGAC CAGCCTGACC AACATGGAGA AACCCCATCT
23543
CTACTAAAAA TACAAAATTA GCTGGGCGTG GTGGCATATG CCTGTAATCC CAGCTACTCG
23603
GGAGGCTGAG GCAGGAGAAT CTTTTGAACC CGGGAGGCAG AGGTTGCGAT GAGCCTAGAT
23663
CGTGCCATTG CACTCCAGCC TGGGCAACAA GAGCAAAACT CGGTCTCAAA AAAAAAAAAA
23723
AAAAAGTGAA ATTAACCAAA GGCATTAGCT TAATAATTTA ATACTGTTTT TAAGTAGGGC
23783
GGGGGGTGGC TGGAAGAGAT CTGTGTAAAT GAGGGAATCT GACATTTAAG CTTCATCAGC
23843
ATCATAGCAA ATCTGCTTCT GGAAGGAACT CAATAAATAT TAGTTGGAGG GGGGGAGAGA
23903
GTGAGGGGTG GACTAGGACC AGTTTTAGCC CTTGTCTTTA ATCCCTTTTC CTGCCACTAA
23963
TAAGGATCTT AGCAGTGGTT ATAAAAGTGG CCTAGGTTCT AGATAATAAG ATACAACAGG
24023
CCAGGCACAG TGGCTCATGC CTATAATCCC AGCACTTTGG GAGGGCAAGG CGAGTGTCTC
24083
ACTTGAGATC AGGAGTTCAA GACCAGCCTG GCCAGCATGG CGATACTCTG TCTCTACTAA
24143
AAAAAATACA AAAATTAGCC AGGCATGGTG GCATGCACCT GTAATCCCAG CTACTCGTGA

24203
GCCTGAGGCA GAAGAATCGC TTGAAACCAG GAGGTGTAGG CTGCAGTGAG CTGAGATCGC
24263
ACCACTGCAC TCCAGCCTGG GCGACAGAAT GAGACTTTGT CTCAAAAAAA GAAAAAGATA
24323
CAACAGGCTA CCCTTATGTG CTCACCTTTC ACTGTTGATT ACTAGCTATA AAGTCCTATA
24383
AAGTTCTTTG GTCAAGAACC TTGACAACAC TAAGAGGGAT TTGCTTTGAG AGGTTACTGT
24443
CAGAGTCTGT TTCATATATA TACATATACA TGTATATATG TATCTATATC CAGGCTTGGC
24503
CAGGGTTCCC TCAGACTTTC CAGTGCACTT GGGAGATGTT AGGTCAATAT CAACTTTCCC
24563
TGGATTCAGA TTCAACCCCT TCTGATGTAA AAAAAAAAAA AAAAAAGAAA GAAATCCCTT
24623
TCCCCTTGGA GCACTCAAGT TTCACCAGGT GGGGCTTTCC AAGTTGGGGG TTCTCCAAGG
24683
TCATTGGGAT TGCTTTCACA TCCATTTGCT ATGTACCTTC CCTATGATGG CTGGGAGTGG
24743
TCAACATCAA AACTAGGAAA GCTACTGCCC AAGGATGTCC TTACCTCTAT TCTGAAATGT
24803
GCAATAAGTG TGATTAAAGA GATTGCCTGT TCTACCTATC CACACTCTCG CTTTCAACTG
24863
TAACTTTCTT TTTTTCTTTT TTTCTTTTTT TCTTTTTTTT TGAAACGGAG TCTCGCTCTG
24923
TCGCCCAGGC TAGAGTGCAG TGGCACGATC TCAGCTCACT GCAAGCTCTG CCTCCCGGGT
24983
TCACGCCATT CTCCTGCCTC ACCCTCCCAA GCAGCTGGGA CTACAGGCGC CTGCCACCAT
25043
GCCCAGCTAA TTTTTTGTAT TTTTAGTAGA GACGGGGTTT CACCGTGTTA GCCAGGATGG
25103
TCTCGATCTC CTGAACTTGT GATCCGCCCG CCTCAGCCTC CCAAAGTGCT GGGATTACAG
25163
GCGTGAGCCA TCGCACCCGG CTCAACTGTA ACTTTCTATA CTGGTTCATC TTCCCCTGTA
25223
ATGTTACTAG AGCTTTTGAA GTTTTGGCTA TGGATTATTT CTCATTTATA CATTAGATTT
25283
CAGATTAGTT CCAAATTGAT GCCCACAGCT TAGGGTCTCT TCCTAAATTG TATATTGTAG
25343
ACAGCTGCAG AAGTGGGTGC CAATAGGGGA ACTAGTTTAT ACTTTCATCA ACTTAGGACC
25403
CACACTTGTT GATAAAGAAC AAAGGTCAAG AGTTATGACT ACTGATTCCA CAACTGATTG
25463
AGAAGTTGGA GATAACCCCG TGACCTCTGC CATCCAGAGT CTTTCAGGCA TCTTTGAAGG
25523
ATGAAGAAAT GCTATTTTAA TTTTGGAGGT TTCTCTATCA GTGCTTAGGA TCATGGGAAT
25583
CTGTGCTGCC ATGAGGCCAA AATTAAGTCC AAAACATCTA CTGGTTCCAG GATTAACATG
25643
GAAGAACCTT AGGTGGTGCC CACATGTTCT GATCCATCCT GCAAAATAGA CATGCTGCAC
25703
TAACAGGAAA AGTGCAGGCA GCACTACCAG TTGGATAACC TGCAAGATTA TAGTTTCAAG
25763
TAATCTAACC ATTTCTCACA AGGCCCTATT CTGTGACTGA AACATACAAG AATCTGCATT
25823
TGGCCTTCTA AGGCAGGGCC CAGCCAAGGA GACCATATTC AGGACAGAAA TTCAAGACTA
25883
CTATGGAACT GGAGTGCTTG GCAGGGAAGA CAGAGTCAAG GACTGCCAAC TGAGCCAATA
25943
CAGCAGGCTT ACACAGGAAC CCAGGGCCTA GCCCTACAAC AATTATTGGG TCTATTCACT
26003

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GTAAGTTTTA ATTTCAGGCT CCACTGAAAG AGTAAGCTAA GATTCCTGGC ACTTTCTGTC
26063
TCTCTCACAG TTGGCTCAGA AATGAGAACT GGTCAGGCCA GGCATGGTGG CTTACACCTG
26123
GAATCCCAGC ACTTTGGGAG GCCGAAGTGG GAGGGTCACT TGAGGCCAGG AGTTCAGGAC
26183
CAGCTTAGGC AACAAAGTGA GATACCCCCT GACCCCTTCT CTACAAAAAT AAATTTTAAA
26243
AATTAGCCAA ATGTGGTGGT GTATACTTAC AGTCCCAGCT ACTCAGGAGG CTGAGGCAGG
26303
GGGATTGCTT GAGCCCAGGA ATTCAAGGCT GCAGTGAGCT ATGATTTCAC CACTGCACTT
26363
CTGGCTGGGC AACAGAGCGA GACCCTGTCT CAAAGCAAAA AGAAAAAGAA ACTAGAACTA
26423
GCCTAAGTTT GTGGGAGGAG GTCATCATCG TCTTTAGCCG TGAATGGTTA TTATAGAGGA
26483
CAGAAATTGA CATTAGCCCA AAAAGCTTGT GGTCTTTGCT GGAACTCTAC TTAATCTTGA
26543
GCAAATGTGG ACACCACTCA ATGGGAGAGG AGAGAAGTAA GCTGTTTGAT GTATAGGGGA
26603
AAACTAGAGG CCTGGAACTG AATATGCATC CCATGACAGG GAGAATAGGA GATTCGGAGT
26663
TAAGAAGGAG AGGAGGTCAG TACTGCTGTT CAGAGATTTT TTTTATGTAA CTCTTGAGAA
26723
GCAAAACTAC TTTTGTTCTG TTTGGTAATA TACTTCAAAA CAAACTTCAT ATATTCAAAT
26783
TGTTCATGTC CTGAAATAAT TAGGTAATGT TTTTTTCTCT ATAG GAA ATG AAT CCT
26839
                                                    Glu Met Asn Pro
                                                    85
CCT GAT AAC ATC AAG GAT ACA AAA AGT GAC ATC ATA TTC TTT CAG AGA
26887
Pro Asp Asn Ile Lys Asp Thr Lys Ser Asp Ile Ile Phe Phe Gln Arg
    90                  95                  100
AGT GTC CCA GGA CAT GAT AAT AAG ATG CAA TTT GAA TCT TCA TCA TAC
26935
Ser Val Pro Gly His Asp Asn Lys Met Gln Phe Glu Ser Ser Ser Tyr
105                 110                 115                 120
GAA GGA TAC TTT CTA GCT TGT GAA AAA GAG AGA GAC CTT TTT AAA CTC
26983
Glu Gly Tyr Phe Leu Ala Cys Glu Lys Glu Arg Asp Leu Phe Lys Leu
                125                 130                 135
ATT TTG AAA AAA GAG GAT GAA TTG GGG GAT AGA TCT ATA ATG TTC ACT
27031
Ile Leu Lys Lys Glu Asp Glu Leu Gly Asp Arg Ser Ile Met Phe Thr
            140                 145                 150
GTT CAA AAC GAA GAC T AGCTATTAAA ATTTCATGCC GGGCGCAGTG GCTCACGCCT
27087
Val Gln Asn Glu Asp
        155
GTAATCCCAG CCCTTTGGGA GGCTGAGGCG GGCAGATCAC CAGAGGTCAG GTGTTCAAGA
27147
CCAGCCTGAC CAACATGGTG AAACCTCATC TCTACTAAAA ATACAAAAAA TTAGCTGAGT
27207
GTAGTGACCC ATGCCCTCAA TCCCAGCTAC TCAAGAGGCT GAGGCAGGAG AATCACTTGC
27267
ACTCCGGAGG TGGAGGTTGT GGTGAGCCGA GATTGCACCA TTGCGCTCTA GCCTGGGCAA
27327
CAACAGCAAA ACTCCATCTC AAAAAATAAA ATAAATAAAT AAACAAATAA AAAATTCATA
27387
ATGTGAACTG TCTGAATTTT TATGTTTAGA AAGATTATGA GATTATTAGT CTATAATTGT
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27447
AATGGTGAAA TAAAATAAAT ACCAGTCTTG AAAAACATCA TTAAGAAATG AATGAACTTT
27507
CACAAAAGCA AACAAACAGA CTTTCCCTTA TTTAAGTGAA TAAAATAAAA TAAAATAAAA
27567
TAATGTTTAA AAAATTCATA GTTTGAAAAC ATTCTACATT GTTAATTGGC ATATTAATTA
27627
TACTTAATAT AATTATTTTT AAATCTTTTG GGTTATTAGT CCTAATGACA AAAGATATTG
27687
ATATTTGAAC TTTCTAATTT TTAAGAATAT CGTTAAACCA TCAATATTTT TATAAGGAGG
27747
CCACTTCACT TGACAAATTT CTGAATTTCC TCCAAAGTCA GTATATTTTT AAAATTCAGT
27807
TTGATCCTGA ATCCAGCAAT ATATAAAAGG GATTATATAC TCTGGCCAAC TGACATTCAT
27867
CCTAGGAATG CAAAGATGGT TTAATATCCT AAAATCAATT AACATAACAT ACTATATTAA
27927
TAAAGTATCA AAACAGTATT CTCATCTTTT TTTCTTTTTT CACAATTCCT TGGTTACACT
27987
ATCATCTCAA TAGATGCAGA AAAAGCATTT GACAAAATCC AATTCATAAT AAAAATTCTC
28047
AAACTTGAAA GAGAACATCA TAAAGGCATC TATGAAAAAC CTACAGCTAA TATCATACTT
28107
AACGATGAAA AACTGAATTA TTTTACCCTA AGATCAAGAA TAATGCAAGC ATGTCAGCTC
28167
TTGCAACTTC TATTCAACAT TGTACTGGAG GTTCTAGCCA GAGCAACCAT ACAATAAATA
28227
AAAATAAAAG GCACCCAGAT TAGAAAGGAA GTCTTTATTT GCAGACAACA TGGTTCTTTA
28287
TGCAGAAAAC CGTCAGGAAT ACACACACAT GTTAGAACTA ATAAGTTCAG CAAGGTTGCA
28347
GGTTGCAATA TCAATATGCA AAAATACATT GAAGGCTGGG CTCAGTGGAG ATGGCATGTA
28407
CCTTTCGTCC CAGCTACTTG GGAGGCTGAG GTAGGAGGAT CACTTGAGGT GAGGAGTTTG
28467
AGGCTATAGT GCAATGTGAT CTTGCCTGTG AATAGCCACT GCACTCGAGC CTAGGCAACA
28527
AAGTGAGACC CCGTCTCCAA AAAAAAAAAT GGTATATTGG TATTTCTGTA TATGAACAAT
28587
GAATGATCTG AAAACAAGAA AATTCCATTC ACGATGGTAT TAAAAAAATA AAATACAAAT
28647
AAATTTAGCA AAATAATTAT AAAACTTGTA CATCGAAAAT TTCAAAGCAC TCTGAGGGAA
28707
ATTAAAGATG ATCTAAATAA TTGGAGAGAC ACTCTATGAT CACTGATTGG AAAATTCATT
28767
CAATATTGTT AAGATAACAA TTGTCCCCAA ATTGATGCAT GCATTCAATT TAGTCTTCAT
28827
CAAAATTCCA GCAGGGTTTT TGCAGAAATT GACAAGCTGT ACCCAAAATG TATATGGAAA
28887
TGAAAAGACC CAGAAGAGCA AATAATTTTT TAAAAACAAA GTTGGAAAAC TTTTACTTCC
28947
TAATTTTAAA ACTTACTATA AACCTAAAGT TATCAAGACC ATTTAGT
28994

(16) INFORMATION FOR SEQ ID NO:15:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH:10 amino acids
(B) TYPE:amino acid
(D) TOPOLOGY:linear
(ii) MOLECULE TYPE:peptide

(v) FRAGMENT TYPE:N-terminal fragment
(xi) SEQUENCE DESCRIPTION:SEQ ID NO:15:

```
Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser
1               5                  10
```

Claims

1. A genomic DNA, which encodes a polypeptide capable of inducing the production of interferon-γ by immunocompetent cells; said polypeptide possessing an amino acid sequence given in SEQ ID NO:1 (where the symbol "Xaa" means "isoleucine" or "threonine") or one of functional equivalents thereof;

SEQ ID NO: 1:

```
Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser Val Ile Arg Asn Leu Asn
1               5                  10                  15
Asp Gln Val Leu Phe Ile Asp Gln Gly Asn Arg Pro Leu Phe Glu Asp
            20                  25                  30
Met Thr Asp Ser Asp Cys Arg Asp Asn Ala Pro Arg Thr Ile Phe Ile
        35                  40                  45
Ile Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met Ala Val Thr Ile
    50                  55                  60
Ser Val Lys Cys Glu Lys Ile Ser Xaa Leu Ser Cys Glu Asn Lys Ile
65                  70                  75                  80
Ile Ser Phe Lys Glu Met Asn Pro Pro Asp Asn Ile Lys Asp Thr Lys
                85                  90                  95
Ser Asp Ile Ile Phe Phe Gln Arg Ser Val Pro Gly His Asp Asn Lys
            100                 105                 110
Met Gln Phe Glu Ser Ser Ser Tyr Glu Gly Tyr Phe Leu Ala Cys Glu
        115                 120                 125
Lys Glu Arg Asp Leu Phe Lys Leu Ile Leu Lys Lys Glu Asp Glu Leu
    130                 135                 140
Gly Asp Arg Ser Ile Met Phe Thr Val Gln Asn Glu Asp
145                 150                 155.
```

2. The genomic DNA of claim 1, which comprises two or more exons; each of the exons possessing a part of nucleotide sequence given in SEQ ID NO:2;

SEQ ID NO: 2:

```
GCCTGGACAG TCAGCAAGGA ATTGTCTCCC AGTGCATTTT GCCCTCCTGG CTGCCAACTC   60
TGGCTGCTAA AGCGGCTGCC ACCTGCTGCA GTCTACACAG CTTCGGGAAG AGGAAAGGAA  120
CCTCAGACCT TCCAGATCGC TTCCTCTCGC AACAAACTAT TTGTCGCAGG AATAAAG     177
ATG GCT GCT GAA CCA GTA GAA GAC AAT TGC ATC AAC TTT GTG GCA ATG     225
```

```
Met Ala Ala Glu Pro Val Glu Asp Asn Cys Ile Asn Phe Val Ala Met
    -35                 -30                 -25
AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA GCT GAA GAT GAT GAA AAC   273
Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala Glu Asp Asp Glu Asn
-20                 -15                 -10                 -5
CTG GAA TCA GAT TAC TTT GGC AAG CTT GAA TCT AAA TTA TCA GTC ATA   321
Leu Glu Ser Asp Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser Val Ile
                1                   5                  10
AGA AAT TTG AAT GAC CAA GTT CTC TTC ATT GAC CAA GGA AAT CGG CCT   369
Arg Asn Leu Asn Asp Gln Val Leu Phe Ile Asp Gln Gly Asn Arg Pro
        15                  20                  25
CTA TTT GAA GAT ATG ACT GAT TCT GAC TGT AGA GAT AAT GCA CCC CGG   417
Leu Phe Glu Asp Met Thr Asp Ser Asp Cys Arg Asp Asn Ala Pro Arg
    30                  35                  40
ACC ATA TTT ATT ATA AGT ATG TAT AAA GAT AGC CAG CCT AGA GGT ATG   465
Thr Ile Phe Ile Ile Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met
45                  50                  55                  60
GCT GTA ACT ATC TCT GTG AAG TGT GAG AAA ATT TCA AYT CTC TCC TGT   513
Ala Val Thr Ile Ser Val Lys Cys Glu Lys Ile Ser Xaa Leu Ser Cys
                65                  70                  75
GAG AAC AAA ATT ATT TCC TTT AAG GAA ATG AAT CCT CCT GAT AAC ATC   561
Glu Asn Lys Ile Ile Ser Phe Lys Glu Met Asn Pro Pro Asp Asn Ile
            80                  85                  90
AAG GAT ACA AAA AGT GAC ATC ATA TTC TTT CAG AGA AGT GTC CCA GGA   609
Lys Asp Thr Lys Ser Asp Ile Ile Phe Phe Gln Arg Ser Val Pro Gly
        95                  100                 105
CAT GAT AAT AAG ATG CAA TTT GAA TCT TCA TCA TAC GAA GGA TAC TTT   657
His Asp Asn Lys Met Gln Phe Glu Ser Ser Ser Tyr Glu Gly Tyr Phe
    110                 115                 120
CTA GCT TGT GAA AAA GAG AGA GAC CTT TTT AAA CTC ATT TTG AAA AAA   705
Leu Ala Cys Glu Lys Glu Arg Asp Leu Phe Lys Leu Ile Leu Lys Lys
125                 130                 135                 140
GAG GAT GAA TTG GGG GAT AGA TCT ATA ATG TTC ACT GTT CAA AAC GAA   753
Glu Asp Glu Leu Gly Asp Arg Ser Ile Met Phe Thr Val Gln Asn Glu
                145                 150                 155
GAC TAGCTATTAA AATTTCATGC CGGGCGCAGT GGCTCACGCC TGTAATCCCA        806
Asp
GCCCTTTGGG AGGCTGAGGC GGGCAGATCA CCAGAGGTCA GGTGTTCAAG ACCAGCCTGA  866
CCAACATGGT GAAACCTCAT CTCTACTAAA AATACTAAAA ATTAGCTGAG TGTAGTGACG  926
CATGCCCTCA ATCCCAGCTA CTCAAGAGGC TGAGGCAGGA GAATCACTTG CACTCCGGAG  986
GTAGAGGTTG TGGTGAGCCG AGATTGCACC ATTGCGCTCT AGCCTGGGCA ACAACAGCAA 1046
AACTCCATCT CAAAAAATAA AATAAATAAA TAAACAAATA AAAAATTCAT AATGTGAAAA 1106
AAAAAAAAAA AAAA                                                   1120.
```

3. The genomic DNA of claim 1, which comprises two exons with respective nucleotide sequences given in SEQ ID NOs:3 and 4;

SEQ ID NO: 3:

```
 AA AAC CTG GAA TCA GAT TAC TTT GGC AAG CTT GAA TCT AAA TTA TCA     47
Glu Asn Leu Glu Ser Asp Tyr Phe Gly Lys Leu Glu Ser Lys Leu Ser
```

```
     -5                  1                   5                  10
GTC ATA AGA AAT TTG AAT GAC CAA GTT CTC TTC ATT GAC CAA GGA AAT      95
Val Ile Arg Asn Leu Asn Asp Gln Val Leu Phe Ile Asp Gln Gly Asn
                15                  20                  25
CGG CCT CTA TTT GAA GAT ATG ACT GAT TCT GAC TGT AGA G               135
Arg Pro Leu Phe Glu Asp Met Thr Asp Ser Asp Cys Arg Asp
            30                  35                  40
```

SEQ ID NO: 4:

```
 AT AAT GCA CCC CGG ACC ATA TTT ATT ATA AGT ATG TAT AAA GAT AGC      47
Asp Asn Ala Pro Arg Thr Ile Phe Ile Ile Ser Met Tyr Lys Asp Ser
40                  45                  50                  55
CAG CCT AGA GGT ATG GCT GTA ACT ATC TCT GTG AAG TGT GAG AAA ATT      95
Gln Pro Arg Gly Met Ala Val Thr Ile Ser Val Lys Cys Glu Lys Ile
                60                  65                  70
TCA ACT CTC TCC TGT GAG AAC AAA ATT ATT TCC TTT AAG                 134
Ser Thr Leu Ser Cys Glu Asn Lys Ile Ile Ser Phe Lys
            80                  85.
```

4. The genomic DNA of claim 1, which comprises two exons with respective nucleotide sequences given in SEQ ID NOs:5 and 6;

SEQ ID NO: 5:

```
GAATAAAG ATG GCT GCT GAA CCA GTA GAA GAC AAT TGC ATC AAC TTT GTG     50
         Met Ala Ala Glu Pro Val Glu Asp Asn Cys Ile Asn Phe Val
             -35                 -30                 -25
GCA ATG AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA G                   87
Ala Met Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala
        -20                 -15                 -10
```

SEQ ID NO: 6:

```
 CT GAA GAT GAT G                                                   12
Ala Glu Asp Asp Glu
-10.
```

5. The genomic DNA of claim 3, which comprises additional two exons with respective nucleotide sequences given in SEQ ID NOs:5 and 6.

6. The genomic DNA of claim 1, which comprises an exon with a part of a nucleotide sequence given in SEQ ID NO:7;

SEQ ID NO: 7:

```
GAA ATG AAT CCT CCT GAT AAC ATC AAG GAT ACA AAA AGT GAC ATC ATA      48
Glu Met Asn Pro Pro Asp Asn Ile Lys Asp Thr Lys Ser Asp Ile Ile
85                  90                  95                  100
TTC TTT CAG AGA AGT GTC CCA GGA CAT GAT AAT AAG ATG CAA TTT GAA      96
Phe Phe Gln Arg Ser Val Pro Gly His Asp Asn Lys Met Gln Phe Glu
                105                 110                 115
TCT TCA TCA TAC GAA GGA TAC TTT CTA GCT TGT GAA AAA GAG AGA GAC     144
Ser Ser Ser Tyr Glu Gly Tyr Phe Leu Ala Cys Glu Lys Glu Arg Asp
            120                 125                 130
CTT TTT AAA CTC ATT TTG AAA AAA GAG GAT GAA TTG GGG GAT AGA TCT     192
Leu Phe Lys Leu Ile Leu Lys Lys Glu Asp Glu Leu Gly Asp Arg Ser
        135                 140                 145
ATA ATG TTC ACT GTT CAA AAC GAA GAC TAGCTAT TAAAATTTCA TGCCGGGCGC   246
Ile Met Phe Thr Val Gln Asn Glu Asp
    150                 155
AGTGGCTCAC GCCTGTAATC CCAGCCCTTT GGGAGGCTGA GGCGGGCAGA TCACCAGAGG   306
TCAGGTGTTC AAGACCAGCC TGACCAACAT GGTGAAACCT CATCTCTACT AAAAATACAA   366
AAAATTAGCT GAGTGTAGTG ACCCATGCCC TCAATCCCAG CTACTCAAGA GGCTGAGGCA   426
GGAGAATCAC TTGCACTCCG GAGGTGGAGG TTGTGGTGAG CCGAGATTGC ACCATTGCGC   486
TCTAGCCTGG GCAACAACAG CAAAACTCCA TCTCAAAAAA TAAAATAAAT AAATAAACAA   546
ATAAAAAATT CATAATGTGA ACTGTCTGAA TTTTTATGTT TAGAAAGATT ATGAGATTAT   606
TAGTCTATAA TTGTAATGGT GAAATAAAAT AAATACCAGT CTTGAAAAAC ATCATTAAGA   666
AATGAATGAA CTTTCACAAA AGCAAACAAA CAGACTTTCC CTTATTTAAG TGAATAAAAT   726
AAAATAAAAT AAAATAATGT TTAAAAAATT CATAGTTTGA AAACATTCTA CATTGTTAAT   786
TGGCATATTA ATTATACTTA ATATAATTAT TTTTAAATCT TTTGGGTTAT TAGTCCTAAT   846
GACAAAAGAT ATTGATATTT GAACTTTCTA ATTTTTAAGA ATATCGTTAA ACCATCAATA   906
TTTTTATAAG GAGGCCACTT CACTTGACAA ATTTCTGAAT TTCCTCCAAA GTCAGTATAT   966
TTTTAAAATT CAGTTTGATC CTGAATCCAG CAATATATAA AAGGGATTAT ATACTCTGGC  1026
CAACTGACAT TCATCCTAGG AATGCAAAGA TGGTTTAATA TCCTAAAATC AATTAACATA  1086
ACATACTATA TTAATAAAGT ATCAAAACAG TATTCTCATC TTTTTTTCTT TTTTCACAAT  1146
TCCTTGGTTA CACTATCATC TCAATAGATG CAGAAAAAGC ATTTGACAAA ATCCAATTCA  1206
TAATAAAAAT TCTCAAACTT GAAAGAGAAC ATCATAAAGG CATCTATGAA AAACCTACAG  1266
CTAATATCAT ACTTAACGAT GAAAAACTGA ATTATTTTAC CCTAAGATCA AGAATAATGC  1326
AAGCATGTCA GCTCTTGCAA CTTCTATTCA ACATTGTACT GGAGGTTCTA GCCAGAGCAA  1386
CCATACAATA AATAAAAATA AAAGGCACCC AGATTAGAAA GGAAGTCTTT ATTTGCAGAC  1446
AACATGGTTC TTTATGCAGA AAACCGTCAG GAATACACAC ACATGTTAGA ACTAATAAGT  1506
TCAGCAAGGT TGCAGGTTGC AATATCAATA TGCAAAAATA CATTGAAGGC TGGGCTCAGT  1566
GGAGATGGCA TGTACCTTTC GTCCCAGCTA CTTGGGAGGC TGAGGTAGGA GGATCACTTG  1626
AGGTGAGGAG TTTGAGGCTA TAGTGCAATG TGATCTTGCC TGTGAATAGC CACTGCACTC  1686
GAGCCTAGGC AACAAAGTGA GACCCCGTCT CCAAAAAAAA AAATGGTATA TTGGTATTTC  1746
TGTATATGAA CAATGAATGA TCTGAAAACA AGAAAATTCC ATTCACGATG GTATTAAAAA  1806
AATAAAATAC AAATAAATTT AGCAAAATAA TTATAAAACT TGTACATCGA AAATTTCAAA  1866
GCACTCTGAG GGAAATTAAA GATGATCTAA ATAATTGGAG AGACACTCTA TGATCACTGA  1926
TTGGAAAATT CATTCAATAT TGTTAAGATA ACAATTGTCC CCAAATTGAT GCATGCATTC  1986
AATTTAGTCT TCATCAAAAT TCCAGCAGGG TTTTTGCAGA AATTGACAAG CTGTACCCAA  2046
AATGTATATG GAAATGAAAA GACCCAGAAG AGCAAATAAT TTTTTAAAAA CAAAGTTGGA  2106
AAACTTTTAC TTCCTAATTT TAAAACTTAC TATAAACCTA AAGTTATCAA GACCATTTAG  2166
T                                                                  2167.
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7. The genomic DNA of claim 3, which comprises additional one exon with a part of a nucleotide sequence given in SEQ ID NO:7.

8. The genomic DNA of claim 5, which comprises additional one exon with a part of a nucleotide sequence given in SEQ ID NO:7.

9. The genomic DNA of claim 1, which comprises two introns with respective nucleotide sequences given in SEQ ID NOs:8 and 9;

SEQ ID NO: 8:

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GTATTTTTTT TAATTCGCAA ACATAGAAAT GACTAGCTAC TTCTTCCCAT TCTGTTTTAC   60
TGCTTACATT GTTCCGTGCT AGTCCCAATC CTCAGATGAA AAGTCACAGG AGTGACAATA  120
ATTTCACTTA CAGGAAACTT TATAAGGCAT CCACGTTTTT TAGTTGGGGT AAAAAATTGG  180
ATACAATAAG ACATTGCTAG GGGTCATGCC TCTCTGAGCC TGCCTTTGAA TCACCAATCC  240
CTTTATTGTG ATTGCATTAA CTGTTTAAAA CCTCTATAGT TGGATGCTTA ATCCCTGCTT  300
GTTACAGCTG AAAATGCTGA TAGTTTACCA GGTGTGGTGG CATCTATCTG TAATCCTAGC  360
TACTTGGGAG GCTCAAGCAG GAGGATTGCT TGAGGCCAGG ACTTTGAGGC TGTAGTACAC  420
TGTGATCGTA CCTGTGAATA GCCACTGCAC TCCAGCCTGG GTGATATACA GACCTTGTCT  480
CTAAAATTAA AAAAAAAAAA AAAAAAAACC TTAGGAAAGG AAATTGATCA AGTCTACTGT  540
GCCTTCCAAA ACATGAATTC CAAATATCAA AGTTAGGCTG AGTTGAAGCA GTGAATGTGC  600
ATTCTTTAAA AATACTGAAT ACTTACCTTA ACATATATTT TAAATATTTT ATTTAGCATT  660
TAAAAGTTAA AAACAATCTT TTAGAATTCA TATCTTTAAA ATACTCAAAA AAGTTGCAGC  720
GTGTGTGTTG TAATACACAT TAAACTGTGG GGTTGTTTGT TTGTTTGAGA TGCAGTTTCA  780
CTCTGTCACC CAGGCTGAAG TGCAGTGCAG TGCAGTGGTG TGATCTCGGC TCACTACAAC  840
CTCCACCTCC CACGTTCAAG CGATTCTCAT GCCTCAGTCT CCCGAGTAGG TGGGATTACA  900
GGCATGCACC ACTTACACCC GGCTAATTTT TGTATTTTTA GTAGAGCTGG GGTTTCACCA  960
TGTTGGCCAG GCTGGTCTCA AACCCCTAAC CTCAAGTGAT CTGCCTGCCT CAGCCTCCCA 1020
AACAAACAAA CAACCCCACA GTTTAATATG TGTTACAACA CACATGCTGC AACTTTTATG 1080
AGTATTTTAA TGATATAGAT TATAAAAGGT TGTTTTTAAC TTTTAAATGC TGGGATTACA 1140
GGCATGAGCC ACTGTGCCAG GCCTGAACTG TGTTTTTAAA AATGTCTGAC CAGCTGTACA 1200
TAGTCTCCTG CAGACTGGCC AAGTCTCAAA GTGGGAACAG GTGTATTAAG GACTATCCTT 1260
TGGTTAAATT TCCGCAAATG TTCCTGTGCA AGAATTCTTC TAACTAGAGT TCTCATTTAT 1320
TATATTTATT TCAG                                                   1334
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SEQ ID NO: 9:

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CAACCTTTTG TCTAGAACAG AAATAACAAG AAGCAGAGAA CCATTAAAGT GAATACTTAC  180
TAAAAATTAT CAAACTCTTT ACCTATTGTG ATAATGATGG TTTTTCTGAG CCTGTCACAG  240
GGGAAGAGGA GATACAACAC TTGTTTTATG ACCTGCATCT CCTGAACAAT CAGTCTTTAT  300
ACAAATAATA ATGTAGAATA CATATGTGAG TTATACATTT AAGAATAACA TGTGACTTTC  360
CAGAATGAGT TCTGCTATGA AGAATGAAGC TAATTATCCT TCTATATTTC TACACCTTTG  420
TAAATTATGA TAATATTTTA ATCCCTAGTT GTTTTGTTGC TGATCCTTAG CCTAAGTCTT  480
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AGACACAAGC TTCAGCTTCC AGTTGATGTA TGTTATTTTT AATGTTAATC TAATTGAATA  540
AAAGTTATGA GATCAGCTGT AAAAGTAATG CTATAATTAT CTTCAAGCCA GGTATAAAGT  600
ATTTCTGGCC TCTACTTTTT CTCTATTATT CTCCATTATT ATTCTCTATT ATTTTTCTCT  660
ATTTCCTCCA TTATTGTTAG ATAAACCACA ATTAACTATA GCTACAGACT GAGCCAGTAA  720
GAGTAGCCAG GGATGCTTAC AAATTGGCAA TGCTTCAGAG GAGAATTCCA TGTCATGAAG  780
ACTCTTTTTG AGTGGAGATT TGCCAATAAA TATCCGCTTT CATGCCCACC CAGTCCCCAC  840
TGAAAGACAG TTAGGATATG ACCTTAGTGA AGGTACCAAG GGGCAACTTG GTAGGGAGAA  900
AAAAGCCACT CTAAAATATA ATCCAAGTAA GAACAGTGCA TATGCAACAG ATACAGCCCC  960
CAGACAAATC CCTCAGCTAT CTCCCTCCAA CCAGAGTGCC ACCCCTTCAG GTGACAATTT 1020
GGACTCCCCA TTCTAGACCT GACAGGCAGC TTAGTTATCA AAATAGCATA AGAGGCCTGG 1080
GATGGAAGGG TAGGGTGGAA AGGGTTAAGC ATGCTGTTAC TGAACAACAT AATTAGAAGG 1140
GAAGGAGATG GCCAAGCTCA AGCTATGTGG GATAGAGGAA AACTCAGCTG CAGAGGCAGA 1200
TTCAGAAACT GGGATAAGTC CGAACCTACA GGTGGATTCT TGTTGAGGGA GACTGGTGAA 1260
AATGTTAAGA AGATGGAAAT AATGCTTGGC ACTTAGTAGG AACTGGGCAA ATCCATATTT 1320
GGGGGAGCCT GAAGTTTATT CAATTTTGAT GGCCCTTTTA AATAAAAAGA ATGTGGCTGG 1380
GCGTGGTGGC TCACACCTGT AATCCCAGCA CTTTGGGAGG CCGAGGGGGG CGGATCACCT 1440
GAAGTCAGGA GTTCAAGACC AGCCTGACCA ACATGGAGAA ACCCCATCTC TACTAAAAAT 1500
ACAAAATTAG CTGGGCGTGG TGGCATATGC CTGTAATCCC AGCTACTCGG GAGGCTGAGG 1560
CAGGAGAATC TTTTGAACCC GGGAGGCAGA GGTTGCGATG AGCCTAGATC GTGCCATTGC 1620
ACTCCAGCCT GGGCAACAAG AGCAAAACTC GGTCTCAAAA AAAAAAAAAA AAAAGTGAAA 1680
TTAACCAAAG GCATTAGCTT AATAATTTAA TACTGTTTTT AAGTAGGGCG GGGGGTGGCT 1740
GGAAGAGATC TGTGTAAATG AGGGAATCTG ACATTTAAGC TTCATCAGCA TCATAGCAAA 1800
TCTGCTTCTG GAAGGAACTC AATAAATATT AGTTGGAGGG GGGGAGAGAG TGAGGGGTGG 1860
ACTAGGACCA GTTTTAGCCC TTGTCTTTAA TCCCTTTTCC TGCCACTAAT AAGGATCTTA 1920
GCAGTGGTTA TAAAAGTGGC CTAGGTTCTA GATAATAAGA TACAACAGGC CAGGCACAGT 1980
GGCTCATGCC TATAATCCCA GCACTTTGGG AGGGCAAGGC GAGTGTCTCA CTTGAGATCA 2040
GGAGTTCAAG ACCAGCCTGG CCAGCATGGC GATACTCTGT CTCTACTAAA AAAAATACAA 2100
AAATTAGCCA GGCATGGTGG CATGCACCTG TAATCCCAGC TACTCGTGAG CCTGAGGCAG 2160
AAGAATCGCT TGAAACCAGG AGGTGTAGGC TGCAGTGAGC TGAGATCGCA CCACTGCACT 2220
CCAGCCTGGG CGACAGAATG AGACTTTGTC TCAAAAAAAG AAAAAGATAC AACAGGCTAC 2280
CCTTATGTGC TCACCTTTCA CTGTTGATTA CTAGCTATAA AGTCCTATAA AGTTCTTTGG 2340
TCAAGAACCT TGACAACACT AAGAGGGATT TGCTTTGAGA GGTTACTGTC AGAGTCTGTT 2400
TCATATATAT ACATATACAT GTATATATGT ATCTATATCC AGGCTTGGCC AGGGTTCCCT 2460
CAGACTTTCC AGTGCACTTG GGAGATGTTA GGTCAATATC AACTTTCCCT GGATTCAGAT 2520
TCAACCCCTT CTGATGTAAA AAAAAAAAAA AAAAAGAAAG AAATCCCTTT CCCCTTGGAG 2580
CACTCAAGTT TCACCAGGTG GGGCTTTCCA AGTTGGGGGT TCTCCAAGGT CATTGGGATT 2640
GCTTTCACAT CCATTTGCTA TGTACCTTCC CTATGATGGC TGGGAGTGGT CAACATCAAA 2700
ACTAGGAAAG CTACTGCCCA AGGATGTCCT TACCTCTATT CTGAAATGTG CAATAAGTGT 2760
GATTAAAGAG ATTGCCTGTT CTACCTATCC ACACTCTCGC TTTCAACTGT AACTTTCTTT 2820
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TCCTGCCTCA CCCTCCCAAG CAGCTGGGAC TACAGGCGCC TGCCACCATG CCCAGCTAAT 3000
TTTTTGTATT TTTAGTAGAG ACGGGGTTTC ACCGTGTTAG CCAGGATGGT CTCGATCTCC 3060
TGAACTTGTG ATCCGCCCGC CTCAGCCTCC CAAAGTGCTG GGATTACAGG CGTCAGCCAT 3120
CGCACCCGGC TCAACTGTAA CTTTCTATAC TGGTTCATCT TCCCCTGTAA TGTTACTAGA 3180
GCTTTTGAAG TTTTGGCTAT GGATTATTTC TCATTTATAC ATTAGATTTC AGATTAGTTC 3240
CAAATTGATG CCCACAGCTT AGGGTCTCTT CCTAAATTGT ATATTGTAGA CAGCTGCAGA 3300
AGTGGGTGCC AATAGGGGAA CTAGTTTATA CTTTCATCAA CTTAGGACCC ACACTTGTTG 3360
ATAAAGAACA AAGGTCAAGA GTTATGACTA CTGATTCCAC AACTGATTGA GAAGTTGGAG 3420
ATAACCCCGT GACCTCTGCC ATCCAGAGTC TTTCAGGCAT CTTTGAAGGA TGAAGAAATG 3480
CTATTTTAAT TTTGGAGGTT TCTCTATCAG TGCTTAGGAT CATGGGAATC TGTGCTGCCA 3540
TGAGGCCAAA ATTAAGTCCA AAACATCTAC TGGTTCCAGG ATTAACATGG AAGAACCTTA 3600
GGTGGTGCCC ACATGTTCTG ATCCATCCTG CAAAATAGAC ATGCTGCACT AACAGGAAAA 3660
GTGCAGGCAG CACTACCAGT TGGATAACCT GCAAGATTAT AGTTTCAAGT AATCTAACCA 3720
TTTCTCACAA GGCCCTATTC TGTGACTGAA ACATACAAGA ATCTGCATTT GGCCTTCTAA 3780
GGCAGGGCCC AGCCAAGGAG ACCATATTCA GGACAGAAAT TCAAGACTAC TATGGAACTG 3840
GAGTGCTTGG CAGGGAAGAC AGAGTCAAGG ACTGCCAACT GAGCCAATAC AGCAGGCTTA 3900
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CACAGGAACC CAGGGCCTAG CCCTACAACA ATTATTGGGT CTATTCACTG TAAGTTTTAA 3960
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TGGCTCAGAA ATGAGAACTG GTCAGGCCAG GCATGGTGGC TTACACCTGG AATCCCAGCA 4080
CTTTGGGAGG CCGAAGTGGG AGGGTCACTT GAGGCCAGGA GTTCAGGACC AGCTTAGGCA 4140
ACAAAGTGAG ATACCCCCTG ACCCCTTCTC TACAAAAATA AATTTTAAAA ATTAGCCAAA 4200
TGTGGTGGTG TATACTTACA GTCCCAGCTA CTCAGGAGGC TGAGGCAGGG GGATTGCTTG 4260
AGCCCAGGAA TTCAAGGCTG CAGTGAGCTA TGATTTCACC ACTGCACTTC TGGCTGGGCA 4320
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ATTAGCCCAA AAAGCTTGTG GTCTTTGCTG GAACTCTACT TAATCTTGAG CAAATGTGGA 4500
CACCACTCAA TGGGAGAGGA GAGAAGTAAG CTGTTTGATG TATAGGGGAA AACTAGAGGC 4560
CTGGAACTGA ATATGCATCC CATGACAGGG AGAATAGGAG ATTCGGAGTT AAGAAGGAGA 4620
GGAGGTCAGT ACTGCTGTTC AGAGATTTTT TTTATGTAAC TCTTGAGAAG CAAAACTACT 4680
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TGAAATAATT AGGTAATGTT TTTTTCTCTA TAG                              4773.
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10. The genomic DNA of claim 1, which comprises three introns with respective nucleotide sequences of SEQ ID NOs: 10 to 12 as introns;

SEQ ID NO: 10:

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GTAAGAAATA TCATTCCTCT TTATTTGGAA AGTCAGCCAT GGCAATTAGA GGTAAATAAG   60
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CAACATAAGA AATATAAGCA AAGTCAGAGT AGAATTTTTT TCTTTTATCA GATATGGGAG  180
AGTATCACTT TAGAGGAGAG GTTCTCAAAC TTTTTGCTCT CATGTTCCCT TTACACTAAG  240
CACATCACAT GTTAGCATAA GTAACATTTT TAATTAAAAA TAACTATGTA CTTTTTTAAC  300
AACAAAAAAA AGCATAAAGA GTGACACTTT TTTATTTTTA CAAGTGTTTT AACTGGTTTA  360
ATAGAAGCCA TATAGATCTG CTGGATTCTC ATCTGCTTTG CATTCAGACT ACTGCAATAT  420
TGCACAGAAT GCAGCCTCTG GTAAACTCTG TTGTACACTC ATGAGAGAAT GGGTGAAAAA  480
GACAAATTAC GTCTTAGAAT TATTAGAAAT AGCTTTCACT TTAGGAACTC CCTGAGAATT  540
GCTGCTTTAG AGTGGTAAGA TAAATAAGCT TCTCTTTAAA CGGAATCTCA AGACAGAATC  600
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CACCTTTGGA CTGGGCTACA ATTGGATAAT ATAGCATTCC CCGAGATAAT TTTCTCTCAC  720
AATTAAGGAA AGGGCTGAAT AAATATCTCT GTTTGAAGTT GAATAACAAA AATTAGGACC  780
CCCTAAATTT TAGGGCTCCT GAAATTCGTC TTTTTGCCTA TATTCAGCTA CTTTACGTTC  840
TATTAAATCT TCTTTCAGGC CAGGTGCACT AGCTCATGCC TAGAATCTCA GGCAGGCCTG  900
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GAGATTCAAC TCAATTCTCC TTTTCAAACT AGGCCATTTA AACTACATCA GTTCCATTTT 1740
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TTGAGATGGA GCCCTGCTGT GTCACCCAGG CTGGAGTGCA GTGGCACGAA CTCGGCTCAC 2160
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GCCTCTGGCT AGTTGTCAAA GTATTGCATA ACTAAATTTT TATGTCTGTT TTAAAGAACA 2760
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GACCTCTGTC CAATCTTGTT CAATCAGGTC CATTCTTTTG TTCTTGGTGG TGGTGGTGGT 4560
GTTGACAGAG TTTCGCTTTT GCTGCCCAGG CTGAAGTGCA GTGGAGCACT TCACTGCAAC 4620
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CACCACACCC AGCTAATTTT GTGTTTTCAG TAGAGACAGG GTTTCACCAT GTTGGTCAGG 4740
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ACGGTATAAA AATTATAGTT TATATGGGCA AAATGGAAAT AATGTTAACT CTTCCAAATA 5100
GTTTATCTAG AATGACATAA TTTCAAAGCT GTCAGGTCAA ATGAGTTATA AACTGTTAAC 5160
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SEQ ID NO: 11:

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SEQ ID NO: 12:

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ATGAGATTTA ATGTTTATTG TAG                                        3383.
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11. The genomic DNA of claim 9, which comprises additional three introns with respective nucleotide sequences given in SEQ ID NOs:10 to 12.

12. The genomic DNA of claim 1, which has a nucleotide sequence selected from the group consisting of SEQ ID NO: 13 and its complementary sequence;

SEQ ID NO: 13:

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    Met Ala Ala Glu Pro Val Glu Asp Asn Cys Ile Asn Phe Val Ala
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          -35                  -30                  -25
ATG AAA TTT ATT GAC AAT ACG CTT TAC TTT ATA G   GTAAGG CTAATGCCAT      98
Met Lys Phe Ile Asp Asn Thr Leu Tyr Phe Ile Ala
    -20                  -15                  -10
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                                        Ala Glu Asp Asp Glu
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Glu Ser Lys Leu Ser Val Ile Arg Asn Leu Asn Asp Gln Val Leu Phe
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Cys Arg Asp
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                            Asp Asn Ala Pro Arg Thr Ile Phe Ile
                            40                  45
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Ile Ser Met Tyr Lys Asp Ser Gln Pro Arg Gly Met Ala Val Thr Ile
    50                  55                  60
TCT GTG AAG TGT GAG AAA ATT TCA ACT CTC TCC TGT GAG AAC AAA ATT      6439
Ser Val Lys Cys Glu Lys Ile Ser Thr Leu Ser Cys Glu Asn Lys Ile
65                  70                  75                  80
ATT TCC TTT AAG GTAAG ACTGAGCCTT ACTTTGTTTT CAATCATGTT AATATAATCA   6496
Ile Ser Phe Lys
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                                                 Glu Met Asn
                                                 85
CCT CCT GAT AAC ATC AAG GAT ACA AAA AGT GAC ATC ATA TTC TTT CAG    11281
Pro Pro Asp Asn Ile Lys Asp Thr Lys Ser Asp Ile Ile Phe Phe Glu
       90                  95                  100
AGA AGT GTC CCA GGA CAT GAT AAT AAG ATG CAA TTT GAA TCT TCA TCA    11329
Arg Ser Val Pro Gly His Asp Asn Lys Met Gln Phe Glu Ser Ser Ser
   105                 110                 115
TAC GAA GGA TAC TTT CTA GCT TGT GAA AAA GAG AGA GAC CTT TTT AAA    11377
Tyr Glu Gly Tyr Phe Leu Ala Cys Glu Lys Glu Arg Asp Leu Phe Lys
120                 125                 130                 135
CTC ATT TTG AAA AAA GAG GAT GAA TTG GGG GAT AGA TCT ATA ATG TTC    11425
Leu Ile Leu Lys Lys Glu Asp Glu Leu Gly Asp Arg Ser Ile Met Phe
                140                 145                 150
ACT GTT CAA AAC GAA GAC TAGCTATTAA AATTTCATGC C                    11464
Thr Val Gln Asn Glu Asp
           155.
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13. The genomic DNA of claim 1, which has a nucleotide sequence selected from the group consisting of SEQ ID NO: 14 and its complementary sequence;

SEQ ID NO: 14:

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TATAAAATAG TCCGGAAATT TCAGAGAAAG ATGAATCTGA TTTTCCAAGA GAGGACAGCT    240
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14. The genomic DNA of claim 1, which is derived from human.

15. The genomic DNA of claim 1, which is inserted into an autonomously replicable vector.

16. A transformant derived from a mammalian cell, which contains the genomic DNA claim 1.

17. The transformant of claim 16, which is derived from a cell selected from the group consisting of epithelial, interstitial and hemopoietic cells from mammal.

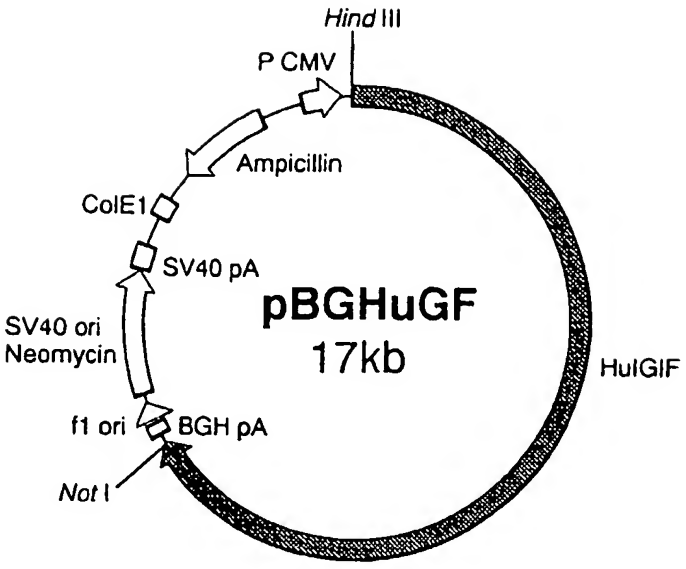
18. A process for preparing a polypeptide, which comprises (a) artificially expressing the DNA of claim 1, and (b) collecting a polypeptide capable of inducing the production of interferon-γ by immunocompetent cells from the resultant mixture.

19. The process of claim 18, wherein the artificial expression of the step (a) comprises a step of culturing the transformant of claim 16.

20. The process of claim 18, wherein the resultant mixture of the step (b) contains a culture of the transformant of claim 16.

21. The process of claim 18, wherein the polypeptide is collected by one or more techniques selected from the group consisting of salting out, dialysis, filtration, concentration, separatory sedimentation, ion-exchange chromatography, gel filtration chromatography, adsorption chromatography, isoelectricpoint chromatography, hydrophobic chromatography, reversed phase chromatography, affinity chromatography, gel electrophoresis and isoelectric focusing.

22. The process of claim 18, wherein the polypeptide is collected by an immunoaffinity chromatography with a monoclonal antibody.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P CMV
Ampicillin
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Neomycin
HuIGIF
f1 ori
BGH pA
Not I

FIG.1